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L5: Entry 217 of 511 File: USPT May 20, 1980

DOCUMENT-IDENTIFIER: US 4204005 A
TITLE: Process for producing fibrous food materials

Detailed Description Text (48):
A mixture consisting of 7 kg of crushed bones of cocks, pigs, and oxen, 2 kg of wheat, degreased soybean, whole grains of soybean and 1 kg of dried okara (residue of soybeans from which soluble matter has been extracted) was stirred to attain uniform distribution and the water content was adjusted to 45% by weight. The resulting material was treated with a colloid mill as in Example 1. Since heating is not contained in the pre-treatment, heating was introduced in the post-treatment as it was in Example 1. Thus, heating was conducted for 5 minutes with steam, followed by dehydration by centrifugation, to obtain fibrous food material.

h e b b g e e e f e e e

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L5: Entry 224 of 511 File: USPT Jan 30, 1979

DOCUMENT-IDENTIFIER: US 4137339 A
TITLE: Method of preparing processed food material from soybean

Abstract Text (1):
A method of preparing a processed food material from soybean comprising soaking whole grains of soybean in water, heating the grains at 80 - 200.degree. C with superheated steam for less than a minute to deactivate the physiologically active substances in the soybean, crushing the grains thus treated with water followed by further subdividing into finer particles with a homogenizer to completely destroy the soybean cells, and adding a protein coagulating agent to precipitate the protein together with fat and fibrin.

Abstract Text (5):
Soybean contains an enzyme which, on being activated, reacts with oil in the soybean grain. The reaction product causes the soybean odor.

Abstract Text (10):
Thus, the present inventors have invented a process for efficiently converting soybean into a processed food material. The features of the present invention lie in the method of preparing a processed food material from soybean comprising soaking whole grains of soybean in water, heating the grains at 80 - 200.degree. C with superheated steam for less than a minute to deactivate the physiologically active substances in the soybean, crushing in cold or hot water followed by further subdividing into finer particles with a homogenizer to completely destroy the soybean cells, adding a protein coagulating agent, such as, acid and salts of calcium to precipitate the protein together with fat and fibrin, the mixture being either curded to obtain a pasty product or dehydrated by freezing or heating to obtain a dried powdery product.

Abstract Text (14):
From the crushed soybean grains the outer skins and cell membranes are separated from the soybean milk by filtration or any other suitable method, forming a solid residue called "okara".

Abstract Text (15):
When soybean curd, which is called "tofu" or frozen dried soybean curd is prepared, the amount of the components utilized remains less than 50% of the total soybean grains, and the solid residue contains more than 20% of the protein and more than 6% of the fat oil. To improve the yield of protein, fat, and oil, the solid residue is further crushed in cold or hot water and then subdivided with a homogenizer under a high pressure greater than 100 kg/cm.sup.2 to destroy any cells remaining in the residue.

Detailed Description Text (3):
One kilogram of soybean grains was washed with water and soaked for 16 hours in water at room temperature to yield about 2.3 kg of swollen soybean grains. The soybean was then placed in a high pressure tank, to which high pressure steam at 6 kg/cm.sup.2 (by gauge) was introduced and the drains as well as most of the air inside the tank were removed. The inside pressure was then raised to 6 kg/cm.sup.2 (by gauge) with the valve closed, and the pressure was maintained for 30 seconds. Then the steam was stopped and the pressure released, and the soybean was taken out

and cooled. Water was removed from the soybean grains which were crushed together with 6 kg of cold or hot water into small granules of less than 0.5 mm diameter. A slurry produced by the crushing was homogenized by one or more treatments in a high pressure homogenizer (Manton-Gaulin Co.) under a pressure exceeding 100 kg/cm.sup.2, to destroy the tissues of soybean. In this treatment cells were broken and the extraction became more complete. The liquid was heated to 75.degree. C, and calcium chloride was added under stirring until a concentration of 0.02 N was reached, to coagulate protein which was then transferred into a frame to make curd from it. The supernatant liquid was removed and the curd was washed with water. Otherwise the coagulum was filtered off with a filtering cloth and further dried by centrifugation with a basket type centrifuge. The yield was about 2.3 kg of coagulum of protein containing 65% of water. This was a processed food material produced from soybean which was free from soybean odor and tasted good. The processed food material could be eaten without further treatment. However, when it was ground with a colloid mill, a pasty product having a fine texture and smooth touch was obtained, of which the composition is as follows:

Detailed Description Text (7):
A liquid containing very fine particles produced in the same process as in Examples 1 and 2 from whole grains of soybean was kept at 37.degree. C, to which about 2% of a starter was added. The starter is selected from the group consisting of lactobacillus vulgaris, lactobacillus thermophyllus, and streptococcus lactis which were cultured as starter for the lactic acid fermentation. Lactic acid was formed during a 4 to 6 hour standing period. Coagulation followed and the coagulum was broken into pieces as large as soybean grains. Water was removed from the grains by mere standing or pressing, to reduce the water content below 65%. This was ground with a colloid mill. Otherwise removal of water was effected by freezing denaturation, and a paste was produced by kneading with a kneader. This paste was dried into a powder by blowing with hot air or by spraying. The components and their contents in the dried product were as follows:

CLAIMS:

1. A method of preparing a food material from soybean comprising swelling the beans by soaking whole grains of soybean in water, heating the swollen grains at 80 to 200.degree. C with superheated steam for less than a minute to deactivate the enzyme and tripsin inhibiting substances therein, crushing the grains thus treated in sufficient amount of water to dissolve flatulence producing saccharides followed by further subdividing into finer particles with a homogenizer to completely destroy the soybean cells and adding a protein coagulating agent in amounts sufficient to precipitate the protein together with fat and fibrin.

L3 ANSWER 1 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1999:281648 BIOSIS
DN PREV199900281648
TI Identification of flavonoid diglycosides in yellow lupin (Lupinus luteus L.) with mass spectrometric techniques.
AU Franski, Rafal; Bednarek, Pawel; Wojtaszek, Przemyslaw; Stobiecki, Maciej [Reprint author]
CS Polish Academy of Sciences, Institute for Bioorganic Chemistry, Noskowskiego 12/14, 61-704, Poznan, Poland
SO Journal of Mass Spectrometry, (**May, 1999**) Vol. 34, No. 5, pp. 486-495. print.
ISSN: 1076-5174.
DT Article
LA English
ED Entered STN: 28 Jul 1999
Last Updated on STN: 28 Jul 1999
AB Various mass spectrometric techniques were used for the structural elucidation of flavonoid glycosides isolated from green parts of yellow lupin plants (Lupinus luteus). A methodological approach is proposed consisting of (1) extraction and purification of flavonoid glycosides from plant tissues, (2) registration of liquid secondary ion mass spectra of intact compounds and (3) gas chromatographic/mass spectrometric analyses of those compounds subjected to several types of chemical modification. On the basis of these data, it was possible to define the molecular mass of the glycosides, the structure of the aglycones, the configuration of sugar moieties and the position of glycosidic linkages between sugars in diglycosides or between **aglycone** and sugar moieties. Analysis of the mass spectrometric data permitted structural identification of four flavonoid diglycosides, where the aglycones had flavone, **isoflavone** and flavonol structures. They were recognized as: apigenin 7-O-(2"-O-rhamnopyranosyl)glucopyranoside (1), genistein 4 ,7-O-diglucopyranoside (2), kaempferol 3-O-(6"-rhamnopyranosyl)glucopyranoside (3) and 3 - or 4 -methylquercetin 3-O-(6"-O-rhamnopyranosyl)glucopyranoside (4). Complete structural evaluation of flavonoid glycosides was possible with only submilligram quantities of samples needed to register various mass spectra. It was not possible, however, to define the anomeric configuration of the C-1 carbons in investigated glycosides.

L3 ANSWER 2 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1998:161280 BIOSIS
DN PREV199800161280
TI HPLC analysis of isoflavonoids and other phenolic agents from foods and from human fluids.
AU Franke, Adrian A. [Reprint author]; Custer, Laurie J.; Wang, Weiqun; Shi, Chen Yang
CS Cancer Res. Cent. Hawaii, 1236 Lauhala Street, Honolulu, HI 96813, USA
SO Proceedings of the Society for Experimental Biology and Medicine, (**March, 1998**) Vol. 217, No. 3, pp. 263-273. print.
CODEN: PSEBAA. ISSN: 0037-9727.
DT Article
LA English
ED Entered STN: 6 Apr 1998
Last Updated on STN: 4 May 1998
AB A fast, precise and selective diode array HPLC method is presented for the extraction and analysis of soy isoflavonoids from foods and from human urine, plasma, and breast milk in support of mechanistic and epidemiologic studies assessing the potential cancer protective role of soya or isoflavones. Solid phase or solvent extraction was chosen for isolation, and enzymatic or acid hydrolysis procedures were used for **aglycone** production depending on the matrix to be analyzed. C-18 reversed-phase HPLC was applied to selectively separate and quantitate daidzein (1), glycitein (3), and genistein (4), including their malonyl (a) and acetyl

(b) esters, and their mammalian metabolites equol (6) and O-desmethylangolensin (7), as well as formononetin (2), biochanin-A (5), and coumestrol (8) using a gradient elution system. UV absorbance scans and authentic standards were applied for identification purposes, additional to fluorometric monitoring, electrochemical detection, and GC/MS analysis after trimethyl silylation. Detection limits of 20-mul injections were found to be 1.09, 0.53, 3.28, and 1.00 pmoles for daidzein, genistein, equol, and O-desmethylangolensin (DMA), respectively, by monitoring at the individual compound's absorption maximum. The proposed method was applied to monitor **isoflavone** levels in soy foods and in human plasma, urine and breast milk after challenge with roasted soybeans. Implications of the presented results on the potential activity of isoflavones to prevent cancer by exposing newborn infants to these agents are discussed.

L3 ANSWER 3 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1997:226810 BIOSIS
DN PREV199799518526
TI Chalcone synthase transcripts are detected in Alfalfa root hairs following inoculation with wild-type Rhizobium meliloti.
AU McKhann, Heather I.; Pavia, Nancy L.; Dixon, Richard A.; Hirsch, Ann M. [Reprint author]
CS Dep. Molecular, Cell Dev. Biology, Los Angeles, CA 90095-1606, USA
SO Molecular Plant-Microbe Interactions, (**1997**) Vol. 10, No. 1, pp. 50-58.
CODEN: MPMIEL. ISSN: 0894-0282.
DT Article
LA English
ED Entered STN: 22 May 1997
Last Updated on STN: 9 Jul 1997
AB Flavonoids are involved in a number of critical events in the interaction between nitrogen-fixing bacteria and legumes. To get a better understanding of the importance of flavonoids in the earliest stages of the alfalfa-Rhizobium meliloti symbiosis, we followed the expression of two chalcone synthase (CHS) gene family members as well as of chalcone isomerase (CHI) and **isoflavone** reductase (IFR) genes. CHS transcripts increased 2 to 4 dpi (days postinoculation) with wild-type rhizobia, but not after inoculation with the heterologous R. leguminosarum bv. trifolii or with an exopolysaccharide (exo) mutant of R. meliloti. CHS transcripts were detected in the root hairs and epidermal cells of the root hair zone, and infrequently in nodule primordia. Insignificant CHI and IFR MRNA accumulation over control levels was observed in response to rhizobial inoculation. The slight increase in CHS transcript accumulation following wild-type R. meliloti inoculation was correlated with an observed increase in root flavonoid content as well as a change in the nod gene-inducing activity of the root exudate. The nod gene-inducing flavonoids exuded from wild-type rhizobia-inoculated roots were identified as 4',7-dihydroxyflavone and 4, 4' dihydroxy-2'-methoxychalcone. Although there was a slight increase over the uninoculated controls in the level of medicarpin-3-O-glucoside 6"-O-malonate (MGM) in extracts of roots inoculated with rhizobia, IFR transcript accumulation was not significantly elevated over that of the controls. Moreover, no medicarpin **aglycone** was detected in the inoculated roots. Thus, although inoculation with wild-type rhizobia triggers some of the genes induced during an interaction between a host and a pathogen, the expression of these genes in the Rhizobium-legume interaction is at a very low level, suggesting that rhizobia have evolved a mechanism(s) to avoid triggering the host's defense responses.

L3 ANSWER 4 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1996:112158 BIOSIS
DN PREV199698684293
TI Absorption and excretion of the soy isoflavone genistein in rats.
AU King, Roger A. [Reprint author]; Broadbent, Jessica L.; Head, Richard J.

[Reprint author]
CS CSIRO Div. Human Nutrition, Adelaide, SA 5000, Australia
SO Journal of Nutrition, (**1996**) Vol. 126, No. 1, pp. 176-182.
CODEN: JONUAI. ISSN: 0022-3166.
DT Article
LA English
ED Entered STN: 12 Mar 1996
Last Updated on STN: 13 Mar 1996
AB Rodent models have been used to study the anticarcinogenic properties of the soy isoflavones, particularly genistein, but there is little information regarding the pharmacokinetics of the absorption and excretion of genistein. In this study, rats were given a single oral dose of genistein (20 mg/kg body weight) or an equivalent dose of its glycone forms, as an **isoflavone**-rich soy extract. Concentrations of genistein were measured in plasma, urine and feces at intervals up to 48 h after dosing. Plasma genistein concentration at 2 h after dosing was 11.0 +- 2.3 mu-mol/L in genistein-treated rats compared with 4.93 +- 0.22 mu-mol/L (P = 0.025) in soy extract-treated rats, but there were no significant differences at 8 h and later times. The mean urinary excretion rate during the first 2 h after dosing was more than 10 times higher in the genistein group compared with the soy extract group (0.27 +- 0.08 mu-mol/h and 0.020 +- 0.011 mu-mol/h, respectively, P = 0.017) but the percentage of dose recovered in urine over 48 h was not different between groups (19.9 +- 2.4% genistein treated; 17.5 +- 1.1% soy extract treated). There were no significant differences between groups in the recovery of genistein in feces (21.9 +- 2.8% and 21.1 +- 2.5% of dose, respectively). Only 6.1 +- 0.9% of the daidzein from the soy extract was recovered in the feces. The results suggest that the extent of absorption of genistein is similar for the glycone and **aglycone** forms. Although higher initial plasma concentrations may be achieved with the **aglycone**, similar long-term concentrations exist for both forms of **isoflavone**.

L3 ANSWER 5 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1995:482408 BIOSIS
DN PREV199598496708
TI Suppression of an isoflavonoid phytoalexin defense response in mycorrhizal alfalfa roots.
AU Volpin, Hanne; Phillips, Donald A.; Okon, Yaacov; Kapulnik, Yoram [Reprint author]
CS Fac. Agric., Hebrew Univ. Jerusalem, Rehovot 76100, Israel
SO Plant Physiology (Rockville), (**1995**) Vol. 108, No. 4, pp. 1449-1454.
CODEN: PLPHAY. ISSN: 0032-0889.
DT Article
LA English
ED Entered STN: 9 Nov 1995
Last Updated on STN: 14 Dec 1995
AB Isoflavonoids and steady-state mRNA levels of phenylalanine ammonia-lyase, chalcone isomerase, and **isoflavone** reductase were followed during a rapid, nearly synchronous infection of alfalfa (Medicago sativa L.) roots by the vesicular arbuscular fungus Glomus intraradices (Schenck and Smith) to test whether previously indicated suppression of the host defense response is regulated by changes in the steady-state mRNA level. Relative amounts of steady-state phenylalanine ammonia-lyase mRNA in the mycorrhizal roots doubled between d 14 and 18 and then immediately declined by 75% to reach and maintain a value lower than the control roots through d 21. Relative levels of chalcone isomerase mRNA in the inoculated roots increased 6-fold between d 14 and 17 and then decreased rapidly to the control level. **Isoflavone** reductase mRNA was not induced by mycorrhizal colonization. High-performance liquid chromatography, proton-nuclear magnetic resonance, and fast atom bombardment-mass spectrometry analyses showed consistent increases in formononetin levels and transient increases in medicarpin-3-O-glycoside

and formononetin conjugates in the inoculated roots when colonization began. As colonization increased, levels of formononetin conjugates declined in mycorrhizal roots below those in uncolonized controls. Medicarpin **aglycone**, an alfalfa phytoalexin normally associated with pathogenic infections, was not detected at any stage. These findings supply detailed evidence that, during early colonization of plant roots by symbiotic Glomus, defense transcripts are induced and then subsequently suppressed.

L3 ANSWER 6 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1995:132311 BIOSIS
DN PREV199598146611
TI Constitutive and elicitation induced metabolism of isoflavones and pterocarpans in chickpea (Cicer arietinum) cell suspension cultures.
AU Barz, Wolfgang; Mackenbrock, Ulrike
CS Inst. Biochem. Biotechnol. Pflanzen, Westfaelische Wilhelms-Univ., Hindenburgplatz 55, D-4400 Muenster, Germany
SO Plant Cell Tissue and Organ Culture, (**1994**) Vol. 38, No. 2-3, pp. 199-211.
CODEN: PTCEDJ. ISSN: 0167-6857.
DT Article
LA English
ED Entered STN: 29 Mar 1995
Last Updated on STN: 23 May 1995
AB Constitutive phenolics of chickpea cell suspension cultures are the isoflavones formononetin and biochanin A, the isoflavanones homoferreirin and cicerin and the pterocarpans medicarpin and maackiain. They accumulate as vacuolar malonylglucosides. The biosynthetic pathways to isoflavones, pterocarpans and malonylglucoside conjugates together with their enzymes are explained. Elicitation of cell cultures leads to pronounced increases in the activities of biosynthetic enzymes with differential effects on the enzymes involved in conjugate metabolism. Low elicitor doses favour pterocarpan conjugate formation whereas high doses lead to pterocarpan **aglycone** accumulation accompanied by vacuolar efflux of formononetin and pterocarpan malonylglucosides. Elicitor-induced changes in enzyme activities and vacuolar efflux of conjugates are prevented by application of 10-3 M concentrations of cinnamic acid. Cinnamate is alternatively metabolized to a glucose ester, a S-glutathionyl conjugate and to cell wall bounds forms; these reactions are intensified by elicitation. **Isoflavone** and pterocarpan biosynthesis and conjugate metabolism as regulated by elicitation and cinnamate is depicted in a metabolic grid to explain the complex regulatory pattern of phenolic accumulation in chickpea cell cultures.

L3 ANSWER 7 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1992:352087 BIOSIS
DN PREV199294044312; BA94:44312
TI DETECTION AND CHARACTERIZATION OF AN S ADENOSYL-L-METHIONINE FLAVONOID 7-O-METHYLTRANSFERASE FROM LEAVES OF PRUNUS-YEDOENSIS MATSUM WITH SPECIAL REFERENCE TO PRUNETRIN FORMATION.
AU ISHIKURA N [Reprint author]; NAKAMURA S; MATO M; YAMAMOTO K
CS DEP BIOLOGICAL SCI, FAC SCI, KUMAMOTO UNIV, KUROKAMI, KUMAMOTO 860, JPN
SO Botanical Magazine Tokyo, (**1992**) Vol. 105, No. 1077, pp. 83-94.
CODEN: BOMZA8. ISSN: 0006-808X.
DT Article
FS BA
LA ENGLISH
ED Entered STN: 29 Jul 1992
Last Updated on STN: 10 Sep 1992
AB An enzyme, S-adenosyl-L-methionine: flavonoid 7-O-methyltransferase (F7OMT, EC 2.1.1.6), catalyzing the transfer of the methyl group from S-adenosyl-L-methionine (SAM) to the 7 position of sophoricoside (5,7,4'-trihydroxyisoflavone 4'-O-glucoside) and some of the other flavonoids, was detected in extracts from leaves of Prunus .times.

yedoensis, and it was partially purified (about 203-fold) by a combination of gel filtration and ion-exchange column chromatographies. F7OMT was isolated as a soluble enzyme with a pH optimum of 7.5 in K-phosphate buffer. The molecular mass of F7OMT, which had an isoelectric point at pH 4.1, was estimated by elution from a column of Sephadex G-100 to be about 36 kDa. The activity of F7OMT was stimulated by 14 mM 2-mercaptoethanol (2-ME) and 1-10 mM K+, but strongly inhibited by 1 mM Cu2+, 1 mM Co2+ and reagents that react with sulfhydryl groups. The apparent Km values for sophoricoside, its **aglycone** genistein (5,7,4'-trihydroxyisoflavone) and quercetin were 1.49, 2.19 and 1.89 .mu.M, respectively. The apparent Km value for SAM as methyl donor was 2.08 mM. The specificity of F7OMT for methyl acceptors was not strict; flavonols, flavanones and flavanonols in addition to isoflavones served as methyl acceptor. An examination of P. .times. yedoensis leaves during spring and autumn showed variations in the activities of F7OMT and UDP-glucose: **isoflavone** 4'-O-glucosyltransferase (I4'GT). The activities of F7OMT and I4'GT increased in enlarging leaf tissues and then markedly declined when the leaves approached maturation. In autumn leaves F7OMT activity was scarcely detected, but a small peak of I4'GT activity was observed during autumnal reddening.

L3 ANSWER 8 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1991:208522 BIOSIS
DN PREV199191111747; BA91:111747
TI ELICITOR-INDUCED FORMATION OF PTEROCARPAN PHYTOALEXINS IN CHICKPEA CICER-ARIETINUM L. CELL SUSPENSION CULTURES FROM CONSTITUTIVE ISOFLAVONE CONJUGATES UPON INHIBITION OF PHENYLALANINE AMMONIA LYASE.
AU MACKENBROCK U [Reprint author]; BARZ W
CS LEHRSTUHL FUER BIOCHEM PFLANZEN, WESTFAELISCHE WILHELMS-UNIV HINDENBURGPLATZ 55, D-4400 MUENSTER, BUNDESREPUBLIK DEUTSCHLAND
SO Zeitschrift fuer Naturforschung Section C Journal of Biosciences, (**1991**) Vol. 46, No. 1-2, pp. 43-50.
ISSN: 0939-5075.
DT Article
FS BA
LA ENGLISH
ED Entered STN: 2 May 1991
Last Updated on STN: 14 Jun 1991
AB After inhibition of phenylalanine ammonia lyase by L-.alpha.-aminooxy-.beta.-phenylpropionic acid, the constitutively formed formononetin 7-O-glucoside-6"-O-malonate is metabolized with the **isoflavone aglycone** being used as an intermediate in the elicitor-induced formation of pterocarpan phytoalexins in chickpea cell suspension cultures. In elicited cultures not treated with the inhibitor phytoalexins are synthesized de novo from phenylalanine. Therefore, in chickpea cells the constitutive **isoflavone** conjugate metabolism and the elicitor-induced pterocarpan formation show metabolic linkage under specific physiological conditions.

L3 ANSWER 9 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1990:52133 BIOSIS
DN PREV199089029497; BA89:29497
TI MINOR ISOFLAVONES FROM THE ROOTS OF PUERARIA-MIRIFICA.
AU INGHAM J L [Reprint author]; TAHARA S; DZIEDZIC S Z
CS DEP FOOD AND SCI, FOOD STUDIES BUILD, UNIV READING, WHITEKNIGHTS, PO BOX 226, READING RG6 2AP, ENGLAND, UK
SO Zeitschrift fuer Naturforschung Section C Journal of Biosciences, (**1989**) Vol. 44, No. 9-10, pp. 724-726.
ISSN: 0939-5075.
DT Article
FS BA
LA ENGLISH
ED Entered STN: 11 Jan 1990
Last Updated on STN: 27 Feb 1990

AB The **isoflavone aglycone** kwakhurin hydrate, and the glycosides genistin (genistein-7-O-glucoside) and puerarin-6"-monoacetate have been isolated from a methanolic extract of Pueraria mirifica roots.

L3 ANSWER 10 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1990:3597 BIOSIS
DN PREV199089003597; BA89:3597
TI MODULATING EFFECT OF PLANT FLAVONOIDS ON THE MUTAGENICITY OF N METHYL-N'-NITRO-N-NITROSOGUANIDINE.
AU FRANCIS A R [Reprint author]; SHETTY T K; BHATTACHARYA R K
CS BIOCHEM DIV, BHABHA ATOMIC RES CENTRE, BOMBAY 400085, INDIA
SO Carcinogenesis (Oxford), (**1989**) Vol. 10, No. 10, pp. 1953-1956.
CODEN: CRNGDP. ISSN: 0143-3334.
DT Article
FS BA
LA ENGLISH
ED Entered STN: 5 Dec 1989
Last Updated on STN: 1 Feb 1990
AB Tests have been carried out with several plant flavonoids to detect their ability to suppress mutagenesis in Salmonella typhimurium strain TA100 NR induced by the direct-acting carcinogen N-methyl-N'-nitro-N-nitrosoguanidine. Among the most effective flavonoids are the **isoflavone**, biochanin A, the flavanone glycoside, naringin, and its **aglycone**, naringenin, and several flavonols, e.g. morin, fisetin, kaempferol, gossypetin and quercetin, including a flavonol glycoside, rutin. In particular, naringin possesses exceptional antimutagenic activity, in as much as, less than half the equimolar amount can reduce the mutagenic potency of this carcinogen by 50%. These flavonoids appear to act either by preventing passage of the carcinogen into bacterial cells or by altering some cellular processes.

L3 ANSWER 11 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1985:401263 BIOSIS
DN PREV198580071255; BA80:71255
TI DEGRADATION OF THE ISOFLAVONE BIOCHANIN A AND ITS GLUCOSIDE CONJUGATES BY ASCOCHYTA-RABIEI.
AU KRAFT B [Reprint author]; BARZ W
CS BIOCHEM PFLANZEN, WESTFAELISCHE WILHELMS-UNIV, D-4400 MUENSTER, FRG
SO Applied and Environmental Microbiology, (**1985**) Vol. 50, No. 1, pp. 45-48.
CODEN: AEMIDF. ISSN: 0099-2240.
DT Article
FS BA
LA ENGLISH
AB Strains of A. rabiei which are pathogenic to chickpea (Cicer arietinum L.) readily catabolized the main chickpea **isoflavone** biochanin A (5,7-dihydroxy-4'-methoxyisoflavone). 3'-Hydroxylation and O-demethylation reactions led to the isoflavones pratensein, genistein and orobol, which were rapidly further degraded. Dihydrogenistein and p-hydroxyphenylacetic acid were also identified as catabolites. Biochanin A-7-O-glucoside was degraded, leading to **aglycone** and pratensein. Biochanin A-7-O-glucoside-6''-O-malonate, the main phenolic constituent of chickpea, was very slowly degraded without subsequent accumulation of catabolites.

L3 ANSWER 12 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1985:263987 BIOSIS
DN PREV198579043983; BA79:43983
TI DEGRADATION OF THE ISOFLAVONE BIOCHANIN A-7-O GLUCOSIDE-6-O-MALONATE AND PHENYLACETIC ACIDS BY FUSARIUM-JAVANICUM.
AU SCHLIEPER D [Reprint author]; KOMOSSA D; BARZ W
CS LEHRSTUHL FUER BIOCHEMIE DER PFLANZEN, WESTFALISCHE WILHELMS-UNIV, HINDENBURGPLATZ 55, D-4400 MUENSTER
SO Zeitschrift fuer Naturforschung Section C Journal of Biosciences, (**1984**) Vol. 39, No. 9-10, pp. 882-887.

ISSN: 0939-5075.
DT Article
FS BA
LA ENGLISH
AB The **isoflavone** conjugate biochanin A-7-O-glucoside-6"-O-malonate is degraded by F. javanicum with an esterase to yield biochanin A-7-O-glucoside which is further cleaved by a glucosidase to the **aglycone**. Biochanin A is funnelled into a known catabolic sequence. Induction of the catabolism of p-methoxyphenylacetic acid is linked to biochanin A degradation; p-hydroxyphenylacetic acid and 3,4-dihydroxyphenylacetic acid degradation is substrate-induced.

L3 ANSWER 13 OF 24 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN
AN 1979:254265 BIOSIS
DN PREV197968056769; BA68:56769
TI CLOVER SECONDARY METABOLITES PART 2 ISOLATION OF 5 4 DI HYDROXY-6 7-METHYLENEDIHYDROXY ISO FLAVONE AND ITS 4-O-BETA-D GLUCOSIDE FROM THE ROOTS OF THE RED CLOVER TRIFOLIUM-PRATENSE.
AU FRAISHTAT P D [Reprint author]; POPRAVKO S A; VUL'FSON N S
CS MM SHEMYAKIN INST BIOORG CHEM, ACAD SCI USSR, MOSCOW, USSR
SO Bioorganicheskaya Khimiya, (**1979**) Vol. 5, No. 2, pp. 228-233.
CODEN: BIKHD7. ISSN: 0132-3423.
DT Article
FS BA
LA RUSSIAN
AB Two compounds were isolated from the methanolic extracts of red clover roots and shown to be 5,4'-dihydroxy-6,7-methylenedioxyisoflavone and its 4'-O-.beta.-D-glucoside. The structure of the **aglycone** was confirmed by counter synthesis. The **isoflavone** manifested growth-inhibiting and antifungal activity suppressing the wheat coleoptile cell elongation induced by IAA.

L3 ANSWER 14 OF 24 CAPLUS COPYRIGHT 2003 ACS on STN
AN 1976:59274 CAPLUS
DN 84:59274
TI Oxidative rearrangement of chalcones by thallium(III) nitrate. IV. Synthesis of dalpatin, fujikinin, glycitein, and of other natural isoflavones
AU Antus, Sandor; Farkas, Lorand; Kardos-Balogh, Zsuzsanna; Nogradi, Mihaly
CS Cent. Res. Inst., Hung. Acad. Sci., Budapest, Hung.
SO Chemische Berichte (**1975**), 108(12), 3883-93
CODEN: CHBEAM; ISSN: 0009-2940
DT Journal
LA German
AB Dalpatin (I, R1 = R4 = H, R2 = Me, R3 = .beta.-D-glucopyranosyl, R5 = MeO), fujikinetin (I, R1 = R3 = R4 = R5 = H, R2 = Me), fujikinin (I, R1 = R4 = R5 = H, R2 = Me, R3 = .beta.-D-glucopyranosyl), isoflavones I (R1 = R2 = R4 = H, R3 = Me, R5 = MeO) and I (R1 = MeO, R2 = R3 = Me, R4 = R5 = H), constituents of Cordyla africana, **isoflavone** I (R1 = R4 = MeO, R2 = R3 = Me, R5 = H), and glycitein (II) were prepd. Dalpatin, e.g. was prepd. by condensation of acetophenone III with benzaldehyde IV to give chalcone V, which was acetylated to VI, and the product oxidatively rearranged with Th(NO3)3 in MeOH to propanone VII. VII was not isolated, but deacetylated with NaOMe and cyclized to the 7-PhCH2O analog of dalpatin, debenzylated to the **aglycone** of dalpatin, which was coupled with acetobromoglucose, and sapond. to give dalpatin. Fujikinin and the isoflavones were similarly prepd.

L3 ANSWER 15 OF 24 CAPLUS COPYRIGHT 2003 ACS on STN
AN 1962:719 CAPLUS
DN 56:719
OREF 56:112g-i,113a-e
TI Spectral studies on flavonoid compounds. II. Isoflavones and flavanones
AU Horowitz, Robert M.; Jurd, Leonard

CS U.S. Dept. of Agr., Albany, CA
SO Journal of Organic Chemistry (**1961**), 26, 2446-9
CODEN: JOCEAH; ISSN: 0022-3263
DT Journal
LA Unavailable
AB cf. CA 52, 7302c. -Spectral changes in the ultraviolet spectra of isoflavones (I) and flavonones (II) on the addn. of NaOAc and AlCl3 showed the presence of free 7-OH and 5-OH groups, resp. Specifically substituted II formed chalcones readily in dil. alkali. The effect of NaOAc and AlCl3 on the spectra of I was shown (no., **isoflavone**, .lambda.max. in m.mu. for I in abs. alc., with NaOAc, and with addn. of 3 drops of 10% aq. AlCl3 given): 1, formononetin, 250, 260, --; 2, osajin, 274, 274, --; 3, genistein, 262, 271, 274; 4, genistin, 262, 262, 273; 5, biochanin A, 261, 271, --; 6, pomiferin, 276, 276, --; 7, sophoricoside, 262, 275, 276; 8, santal, 263, --, 274; 9, 7-O-methylpodospicatin, 265, --, 277; 10, irigenin, 267, 277, --; 11, iridin, 268, 268, --; 12, podospicatin, 263, 275, 273; 13, 5 hydroxy-2',5',6,7-tetramethoxyisoflavone, 262, --, 275. The corresponding changes in the spectra of II were (no., II, .lambda.max. in m.mu. of II in abs. alc., abs. alc. satd. with fused NaOAc, 2.5 ml. abs. alc. treated with 1 drop of 1% NaOH, and abs. alc. satd. with AlCl3.6H2O given): 1, 7-hydroxyflavanone, 277, 338, 338, 277; 2, liquiritigenin, 276, 338, 338, 276; 3, butin, 278, 338, 338, 278; 4, pinocembrin, 291, 329, 329, 312; 5, naringenin, 290, 328, 328, 311; 6, eriodictyol, 289, 328, 328, 310; 7, taxifolin, 291,330, 329, 314; 8, isosakuranetin, 292, 328, 328, 312; 9, 4, 5,7-trihydroxy-7-methoxyflavanone, 287, 287, 289, 309; 10, homoeriodictoyl, 289, 328, 328, 311; 11, hesperetin, 288, 328, 328, 311; 12, 5-hydroxy-3',4',7-triacetoxyflavanone, 274, -, -, 303; 13, poncirin, 283, 283, 285, 308; 14, eriocitrin, 285, 285, 285, 306; 15, hesperidin, 285, 285, 287, 308; 16, neohesperidin, 285, 285, 287, 308; 17, sakuranetin, 287, 287, 424, 310; 18, sakuranin, 281, 281, 428, 281; 19, prunin, 284, 284, 425, 308; 20, naringenin 7-rhamnoglucoside, 284, 284, 428, 308. Ionization of a 7-OH group gave a bathochromic shift of .apprx.10 m.mu. in I (1, 3, 5, 7, 10, and 12) and 35-60 m.mu. in II (1, 2, 3, 4, 5, 6, 7, 8, 10, and 11). No significant changes were observed in compds. lacking a free OH group in the 7-position. The spectrum of II (16) was unaffected by NaOAc, and it was inferred that the sugar groups were attached through the 7-OH group. The main absorption band of I (12) was shifted 12 m.mu. on addn. of NaOAc, as would be expected from the structure assigned by Briggs and Cebalo (CA 53, 21911c). NaOH generally gave the same results with II, except in 4'-hydroxy-7-alkoxy- or 4'-hydroxy-7-glucosidoxyflavanones, which rapidly formed chalcones with broad max. at 400-450 m.mu. in the ionized form. The susceptibility was visualized as the result of increased acidity of the .alpha.-II coupled with ionization of the 4'OH group as shown by II (18), which gave no shift with NaOAc or AlCl3, but formed the chalcone in alkali. After hydrolysis to the **aglycone** (II, 17), a shift with AlCl3 was observed. The principal wavelength of 5-hydroxyisoflavones underwent a remarkably const. bathochromic shift of 11-14 m.mu. on addn. of AlCl3, and that of 5-hydroxyflavanones changed by 20-30 m.mu.. The spectra of the new flavanone glycoside eriocitrin (14) showed a bathochromic shift with AlCl3 and lack of a shift with NaOAc, indicating the presence of a 5-OH group and of a sugar group in the 7-OH position. The presence of free o-dihydroxyl groups was inferred from the instability of the compd. in alk. soln., together with other evidence.

L3 ANSWER 16 OF 24 CAPLUS COPYRIGHT 2003 ACS on STN
AN 1960:113357 CAPLUS
DN 54:113357
OREF 54:21637h-i,21638a
TI The constituents of Pueraria root
AU Shibata, Shoji; Murakami, Takao; Nishikawa, Yoshihiro; Harada, Masatoshi
CS Univ. Tokyo
SO Chemical & Pharmaceutical Bulletin (**1959**), 7, 134-6
CODEN: CPBTAL; ISSN: 0009-2363

DT Journal
LA Unavailable
AB The traditional medicinal use of Pueraria root led to a search for its constituents with (probably) antispasmodic activity. The MeOH ext. from 1 kg. dried Pueraria root yielded 40 g. of the fraction pptd. with basic Pb(OAc)2, and this was subjected to chromatography over Al2O3. Five of the 10 fractions evident on the chromatogram were examd. [fraction, m.p., % yield (calcd. from the wt. of dried Pueraria root), and derivs. given]: b, 320.degree. (decompn.), 0.13, diacetate m. 186.degree., di-Me ether m. 160.degree.; c, 215-17.degree. (decompn.), 0.13, pentaacetate m. 186-7.degree.; e, 187.degree. (decompn.), 2.3, hexaacetate m. 129-30.degree. (decompn.); f, -, 0.07, acetate m. 162-7.degree. (decompn.); and g, amorphous, 0.09. Ultraviolet absorption data are recorded for the acetates of fractions b, c, e, and f, and, for comparison, daidzein di-Me ether, and the triacetates of genistein (I) and apigenin (II). These showed the fractions b, c, e, and f to be similar to I and different from II, and, therefore, to be **isoflavone** rather than flavone derivs. Fraction b was identified as daidzein (III), the **aglycone** of daidzin, by mixed fusion of its di-Me ether with that of III. By tests on mouse gut, III was found to account for the antispasmodic action.

L3 ANSWER 17 OF 24 CAPLUS COPYRIGHT 2003 ACS on STN
AN 1958:114792 CAPLUS
DN 52:114792
OREF 52:20420b-e
TI Physiology of some flavonoids and hydroxycinnamic acids. I. Choice and identification of the constituents to be studied
AU Urban, Rosemarie
CS Univ. Heidelberg, Germany
SO Planta (**1958**), 52, 47-64
CODEN: PLANAB; ISSN: 0032-0935
DT Journal
LA Unavailable
AB cf. Reznik, C.A. 51, 5147e. Caffeic, quinic, and chlorogenic acids were selected for study, based among others upon the studies of Herrmann. (C.A. 50, 17329d), as types of secondary plant constituents not contg. N. To assist in locations of these in chromatograms, rutin and its **aglycone** quercetin and scopolin and its diglucoside were used. In the leaves of Hedera helix considerable amts. of rutin, chlorogenic acid caffeic acid, and scopoline were found. The principal flavonoid of Helianthus annus leaves is not quercimeritrin, but a flavanone or **isoflavone**. Chlorogenic and caffeic acids and scopoline were also found. A characteristic of the leaves of corn is the presence of 2 strongly water-sol. derivs. of cinnamic acid. The principal flavonoid for the maize strain studied was not isoquercetrin, but a flavone. Leaves of wheat contained tricin and 2 other flavones and scopoline. Light changed the quantities, but not the identities of the constituents of ivy, maize, and wheat that were studied. Leaves from the vegetative region of ivy contained only rutin as a flavonol; leaves from the reproductive region had as their principal flavonol constituent a glucoside of campherol. The compds. were isolated as spots on paper chromatograms (cf. Hansel and Horhammer, C.A. 50, 12403d). The spots were cut out and extd. and the exts. examd. spectrophotometrically after conversion of the substance to the Al complex (cf. Horhammer and Hansel, C.A. 51, 6619h). Ultraviolet transmission spectra and Rf values are given.

L3 ANSWER 18 OF 24 DISSABS COPYRIGHT (C) 2003 ProQuest Information and Learning Company; All Rights Reserved on STN
AN 1999:11935 DISSABS Order Number: AARMQ30758
TI EFFECTS OF CITRUS JUICES AND CONSTITUENT FLAVONOIDS ON LIPID AND LIPOPROTEIN METABOLISM: IN VIVO AND IN VITRO STUDIES
AU BORRADAILE, NICA MARIA [M.SC.]; CARROLL, KENNETH K. [adviser]
CS THE UNIVERSITY OF WESTERN ONTARIO (CANADA) (0784)

SO Masters Abstracts International, (**1998**) Vol. 37, No. 1, p. 248.
Order No.: AARMQ30758. 77 pages.
ISBN: 0-612-30758-1.
DT Dissertation
FS MAI
LA English
AB Several reports have shown that dietary flavonoids, such as the soy **isoflavone**, genistein, and the citrus flavanone glycoside, hesperidin, have cholesterol-lowering properties in vivo. In order to determine whether replacing drinking water with either orange juice or grapefruit juice (which contain significant quantities of hesperidin and naringin, respectively) could lower cholesterol, we conducted studies in a rabbit model of endogenous hypercholesterolemia.

To determine whether the changes observed in rabbits were due to flavonoids present in the juices acting directly on the liver, the effects of hesperetin and naringenin (**aglycone** forms of hesperidin and naringin, respectively) on net apoB secretion by HepG2 cells were investigated. The flavonoids dose-dependently reduced net apoB secretion by up to 81% after a 24h exposure. Further studies showed that net apoB secretion by the cells was significantly reduced by 50% after a 4h exposure to 60μg/ml of either hesperetin or naringenin. Since the reduction in net apoB secretion occurred after a brief exposure to the flavonoids, we investigated whether the effects could be (i) due to increased proteasomal degradation of apoB prior to secretion, or (ii) due to altered lipid availability during the early stages of lipoprotein assembly. (Abstract shortened by UMI.)

L3 ANSWER 19 OF 24 EMBASE COPYRIGHT 2003 ELSEVIER INC. ALL RIGHTS RESERVED. on STN
AN 1998411060 EMBASE
TI Soy isoflavone analysis: Quality control and a new internal standard.
AU Song T.; Barua K.; Buseman G.; Murphy P.A.
CS P.A. Murphy, Dept. of Food Sci./Human Nutrition, Iowa State University, Ames, IA 50011, United States. pmurphy@iastate.edu
SO American Journal of Clinical Nutrition, (**1998**) 68/6 SUPPL. (1474S-1479S).
Refs: 17
ISSN: 0002-9165 CODEN: AJCNAC
CY United States
DT Journal; Conference Article
FS 029 Clinical Biochemistry
LA English
SL English
AB Development of a database of the soy **isoflavone** content of foods requires accurate and precise evaluation of different food matrixes. To evaluate accuracy, we estimated recoveries of both internal and external standards in 5 different soyfoods weekly. Standards were evaluated daily for system quality assurance. To evaluate sample precision, we analyzed soybeans and soymilk bimonthly for within-day precision and over 4 d for day-to-day precision. CVs should be .ltoreq.8%. We validated our methods for single and multiple recovery concentrations by using our new internal standard, 2,4,4'- trihydroxydeoxybenzoin, and the external standards daidzein, genistein, and genistin. Concentrations of 12 **isoflavone** isomers, 3 aglycones (daidzein, genistein, and glycitein), and 9 glucosides (daidzin, genistin, glycitin, acetyldaidzin, acetylgenistin, acetylglycitin, malonyldaidzin, malonylgenistin, and malonylglycitin) were measured in a variety of soybeans and soyfoods. The extraction methods used depended on soyfood type. The HPLC conditions for soy **isoflavone** analysis were improved, leading to good separation with a short analysis time (60 min/sample). A data bank of concentration and distribution of isoflavones in different soybean products was assembled. A wide range of **isoflavone** concentrations, from <50 .mu.g/g to >20000 .mu.g/g, was found in different soy products. The glucoside forms are almost twice the molecular weight of the aglycones; reported

isoflavone concentrations should be normalized to the **aglycone** mass (or an isoflavonoid equivalent) rather than a simple sum of all isomers.

L3 ANSWER 20 OF 24 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
AN 1998:863629 SCISEARCH
GA The Genuine Article (R) Number: 136JQ
TI Dietary flavonol quercetin induces chloride secretion in rat colon
AU Cermak R; Follmer U; Wolffram S (Reprint)
CS CHRISTIAN ALBRECHTS UNIV KIEL, INST ANIM NUTR PHYSIOL & METAB, OLSHAUSEN STR 40, D-24098 KIEL, GERMANY (Reprint); CHRISTIAN ALBRECHTS UNIV KIEL, INST ANIM NUTR PHYSIOL & METAB, D-24098 KIEL, GERMANY; UNIV ZURICH, INST VET PHYSIOL, CH-8057 ZURICH, SWITZERLAND
CYA GERMANY; SWITZERLAND
SO AMERICAN JOURNAL OF PHYSIOLOGY-GASTROINTESTINAL AND LIVER PHYSIOLOGY, (**NOV 1998**) Vol. 38, No. 5, pp. G1166-G1172.
Publisher: AMER PHYSIOLOGICAL SOC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814.
ISSN: 0193-1857.
DT Article; Journal
FS LIFE
LA English
REC Reference Count: 26
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB The aim of this study was to investigate the possible effects of the flavonol quercetin, the most abundant dietary flavonoid, on the intestinal mucosa. In vitro experiments were performed with various segments of the rat intestine, using the Ussing chamber technique. Quercetin increased the short-circuit current (I-sc) in the jejunum, ileum, and proximal and distal colon. Additional experiments were performed using preparations of the proximal colon. The maximum effective dose of quercetin was found to be similar to 100 mu M. The quercetin-induced increase in I-sc was inhibited by the Cl- channel blocker 5-nitro-2-(3-phenylpropylamino)benzoic acid. Adding blockers of the Na+-K+-2Cl(-) cotransporter to the serosal compartment diminished the increase of I-sc due to quercetin. Ion substitution and flux measurements indicated that the effect of quercetin was due to electrogenic Cl- and HCO3- secretion. In contrast to the **aglycone**, the quercetin glycoside rutin had no effect. The effect of quercetin on I-sc was additive to the I-sc increase induced by forskolin, but the flavonoid diminished the I-sc evoked by carbachol. The phosphodiesterase inhibitor theophylline blocked the effect of quercetin. Genistein, a related **isoflavone**, did not alter the I-sc evoked by quercetin. These findings demonstrate that the dietary flavonol quercetin induces Cl- secretion and most likely HCO3- secretion in rat small and large intestine. The effects are restricted to the flavonol **aglycone**.

L3 ANSWER 21 OF 24 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
AN 94:267947 SCISEARCH
GA The Genuine Article (R) Number: BA14P
TI SOYBEAN SAPONIN AND ISOFLAVONOIDS - STRUCTURE AND ANTIVIRAL ACTIVITY AGAINST HUMAN-IMMUNODEFICIENCY-VIRUS IN-VITRO
AU OKUBO K (Reprint); KUDOU S; UCHIDA T; YOSHIKI Y; YOSHIKOSHI M; TONOMURA M
CS TOHOKU UNIV, FAC AGR, DEPT APPL BIOL CHEM, AOBA KU, 1-1 TSUTSUMIDORI, SENDAI, MIYAGI 981, JAPAN (Reprint); KANESA CO LTD, TAMAGAWA, AOMORI 030, JAPAN; NESTLE JAPAN LTD, CHUO KU, KOBE 651, JAPAN
CYA JAPAN
SO ACS SYMPOSIUM SERIES, (**1994**) Vol. 546, pp. 330-339.
ISSN: 0097-6156.
DT General Review; Journal
FS AGRI
LA ENGLISH
REC Reference Count: 49
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Nine kinds of **isoflavone** glycosides were isolated from the hypocotyls of soybean seeds. Three were proved to be malonylated soybean isoflavones named 6''-O-malonyldaizin, 6''-O-malonylglycitin and 6''-O-malonylgenistin. Soyasaponins are divided into three groups according to their respective type of **aglycone**, soyasapogenol A, B and E. Bb, major constituent of group B saponins, completely inhibited HIV-induced cytopathic effects and virus-specific antigen expression 6 days after infection at concentration greater than 0.25 mg/ml, but had no direct effect on HIV reverse transcriptase activity. Bb also inhibited HIV-induced cell fusion in the MOLT-4 cell system.

L3 ANSWER 22 OF 24 SCISEARCH COPYRIGHT 2003 THOMSON ISI on STN
AN 91:125201 SCISEARCH
GA The Genuine Article (R) Number: EZ406
TI ELICITOR-INDUCED FORMATION OF PTEROCARPAN PHYTOALEXINS IN CHICKPEA (CICER-ARIETINUM L) CELL-SUSPENSION CULTURES FROM CONSTITUTIVE ISOFLAVONE CONJUGATES UPON INHIBITION OF PHENYLALANINE AMMONIA-LYASE
AU MACKENBROCK U; BARZ W (Reprint)
CS UNIV MUNSTER, LEHRSTUHL BIOCHEM PFLANZEN, HINDENBURGPLATZ 55, W-4400 MUNSTER, GERMANY
CYA GERMANY
SO ZEITSCHRIFT FUR NATURFORSCHUNG C-A JOURNAL OF BIOSCIENCES, (**1991**) Vol. 46, No. 1-2, pp. 43-50.
DT Article; Journal
FS LIFE
LA ENGLISH
REC Reference Count: 40
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB After inhibition of phenylalanine ammonia lyase by L-alpha-aminooxy-beta-phenylpropionic acid, the constitutively formed formononetin 7-O-glucoside-6"-O-malonate is metabolized with the **isoflavone aglycone** being used as an intermediate in the elicitor-induced formation of pterocarpan phytoalexins in chickpea cell suspension cultures. In elicited cultures not treated with the inhibitor phytoalexins are synthesized de novo from phenylalanine. Therefore, in chickpea cells the constitutive **isoflavone** conjugate metabolism and the elicitor-induced pterocarpan formation show metabolic linkage under specific physiological conditions.

L3 ANSWER 23 OF 24 USPATFULL on STN
AN 1998:92194 USPATFULL
TI Process for obtaining malonyl isoflavone glycosides and obtaining isoflavone glycosides or isoflavone aglycons from malonyl isoflavone glycosides
IN Matsuura, Masaru, Noda, Japan
Obata, Akio, Noda, Japan
Tobe, Kouichiro, Noda, Japan
Yamaji, Nobuyuki, Noda, Japan
PA Kikkoman Corporation, Noda, Japan (non-U.S. corporation)
PI US 5789581 19980804 <--
AI US 1996-630347 19960410 (8)
PRAI JP 1995-112705 19950414
DT Utility
FS Granted
EXNAM Primary Examiner: Kight, John; Assistant Examiner: White, Everett
LREP Banner & Witcoff, Ltd.
CLMN Number of Claims: 10
ECL Exemplary Claim: 1
DRWN No Drawings
LN.CNT 412
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
AB A malonylisoflavone glycoside present in soybean is obtained efficiently. In addition, the corresponding **isoflavone** glycoside and **aglycone** are obtained from the malonylisoflavone

glycoside. An aqueous extract of soybean is adsorbed on an adsorbent, and eluted with an aqueous alcohol solution.

L3 ANSWER 24 OF 24 USPATFULL on STN
AN 97:96993 USPATFULL
TI Process for the isolation and purification of isoflavones
IN Zheng, BoLin, Superior, CO, United States
Yegge, John A., Longmont, CO, United States
Bailey, David T., Boulder, CO, United States
Sullivan, James L., Louisville, CO, United States
PA Hauser, Inc., Boulder, CO, United States (U.S. corporation)
PI US 5679806 19971021 <--
AI US 1995-394407 19950224 (8)
DT Utility
FS Granted
EXNAM Primary Examiner: Richter, Johann; Assistant Examiner: Stockton, Laura L.
LREP Chrisman, Bynum & Johnson, P.C., Petersen, Steven C.
CLMN Number of Claims: 35
ECL Exemplary Claim: 1
DRWN 6 Drawing Figure(s); 6 Drawing Page(s)
LN.CNT 762
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
AB The present invention relates to a process for the isolation and purification of isoflavones from a number of different biomass sources. More particularly, the present invention relates to a three-step process whereby a biomass containing isoflavones with a solvent thereby forming an extract that is subsequently fractionated using a reverse phase matrix in combination with a step gradient elution, wherein the resulting fractions eluted from the column contain specific isoflavones that are later crystallized. The purified **isoflavone** glycosides may then be hydrolyzed to their respective **aglycone** .

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First Hit Fwd Refs

L1: Entry 1 of 18 File: USPT Oct 7, 2003

DOCUMENT-IDENTIFIER: US 6630178 B1
TITLE: Composition comprising soy protein, dietary fibres and a phytoestrogen compound and use thereof in the prevention and/or treatment of cardiovascular diseases

Detailed Description Text (17):
Without wishing to be bound by any specific theory it is presently believed that both soluble dietary fibres (working as nutrients) and insoluble dietary fibres (working as bulking agents), in particular from soybean fibres, more particularly soy cotyledon fibres, provide favourable growth conditions for the microflora in the human gut, which makes the microflora more effective in deconjugating isoflavones in the glucoside form to the aglycone form. Isoflavones in the aglycone form are absorbed faster and to a greater extent in the human gut than isoflavones in the glucoside form, and isoflavones in the aglycone form are the biologically more active species with regard to lowering lipid serum levels and reducing atherosclerosis. In view hereof it can be understood that administration of a combination of soy proteins, a high, fixed level of isoflavones and a combination of soluble and insoluble fibres is effective in providing an increased uptake of isoflavones.

CLAIMS:

16. A composition according to claim 12 wherein the isoflavones are in the aglycone form.

h e b b g e e e f e e e f

First Hit Fwd Refs

L1: Entry 2 of 18 File: USPT Aug 19, 2003

DOCUMENT-IDENTIFIER: US 6607757 B2
TITLE: Soya extract, process for its preparation and pharmaceutical composition

Brief Summary Text (6):
According to biomedical literature and epidemiological information published in recent years, principally in relation to populations of the East, which consume soya-based foods to a great extent, the use of these foods to a high degree reduces pre-menopausal and post-menopausal symptoms in women (A. Cassidy, Proceedings of the Nutrition Society, 1996, 55, 339-417). These facts, which still lack a clear scientific basis, are usually ascribed to the isoflavone aglycones genistein, daidzein and glycitein, which are present in the various soya-based foods.

Brief Summary Text (8):
According to further biomedical literature and epidemiological information published in recent years, principally relating to population groups in the East, which consume soya-based foods to a great extent, the use of these foods decreases to a high degree breast cancer in women and cancer of the prostate in men (A. Nomura, B. E., Henderson J. Lee, American Journal of Clinical Nutrition, 1978, 31, 2020-2025; T. Hirayama in Diet, Nutrition and Cancer, 1986 pp. 41-53, Y. Hayashi, M. Nagao, T. Sugimura, S. Takayama, L. Tomatis, L. W. Wattenberg and G. N. Wogan eds. Tokyo: Japanese Scientific Society Press; R. K. Severson, A. M. Y. Nomura, J. S. Grove, G. N. Stemmerman, Cancer Research, 1989, 49,1857-1860). Also, these facts, which still lack a clear scientific basis, are usually ascribed to the isoflavone aglycones genistein, daidzein and glycitein which are present in the various soya-based foods.

First Hit Fwd Refs

L1: Entry 3 of 18 File: USPT Feb 11, 2003

DOCUMENT-IDENTIFIER: US 6517831 B2
TITLE: Product containing healthful component and process for preparing the same

Brief Summary Text (44):
In concrete terms, proteins, isoflavones, saponins and phytic acid, etc., are respectively converted into the health-promoting component such as peptides, isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc., by performing a koji preparation treatment and a hydrolysis treatment on beans used as a raw material, and are thus converted into a state in which these components can be very easily absorbed by the digestive tracts of single-stomached animals, etc. Using the product of the present invention which has such a health-promoting component based on the above-described koji preparation treatment and hydrolysis treatment, health promotion by means of at least one member of the group consisting of liver function improving constituents, cardiac function improving constituents, anti-inflammatory constituents, antifat constituents, antioxidation consititents and antimutagen constituents can be accomplished very effectively.

Brief Summary Text (49):
Since the product and preparation process of the present invention are constructed and act as described above, a health-promoting component can be produced in a product using beans as a raw material, by means of a koji preparation treatment and a hydrolysis treatment. In concrete terms, peptides, isoflavones, saponins and phytic acid, etc. are respectively converted into a health-promoting component such as isoflavone aglycones, saponii aglycones, myo-inositol and Maillard reaction products, etc. and are thus converted into a state which can be very easily absorbed by the digestive tracts of single-stomached animals. In the present invention, furthermore, the intestine-regulating bacteria added following the initiation of the above-described koji preparation treatment but prior to the completion of the hydrolysis treatment propagate so that a health-promoting component consisting of such intestine-regulating bacteria can be included in the product in large amounts, thus making it possible to achieve an intestine-regulating effect in the intestines of single-stomached animals. By using the product of the present invention which contains such a health-promoting component, health promotion by means of at least one member of the group consisting of liver function improving constituents, cardiac function improving constituents, anti-inflammatory constituents, antifat constituents, antioxidation consititents and antimutagen constituents and components which have an intestine-regulating effect in single-stomached animals can be accomplished very effectively. This product can be used directly "as is", or the efficacy can be strengthened by extracting and concentrating the isoflavone aglycones, saponin aglycones, myo-inositol or Maillard reaction products, etc; in addition, this product can also be utilized in applied products which use this product as a raw material, e. g., food products, livestock feeds, pet foods or drug raw materials, etc. and is thus superior in terms of all-purpose utility. Furthermore, miso and soy sauce are foods with a high salt content, since salt is added following the koji preparation step, so that such food products lack all-purpose utility; moreover, the aging period of such food products following the koji preparation step is extremely long, so that there are problems in productivity. However, the product of the present invention is easy to manufacture at a low-cost.

h e b b g e e e f e e e f

Detailed Description Text (9):
Afterward, the mixture is placed in a koji preparation device and held for a prescribed period of time with the initial temperature set at approximately 28 to 30.degree. C. Koji preparation is then performed by fermenting the soybean meal with a low moisture content of 40 wt % by means of koji mold until the enzymes required in order to convert the proteins, isoflavones, saponins and phytic acid, etc., contained in the soybean meal into respective health-promoting components such as peptide, isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc., are produced.

Detailed Description Text (11):
With respect to such cases in which a health-promoting component is produced by this koji preparation step, a case will be described in which the glycosides of isoflavones and saponins are decomposed so that respective isoflavone aglycones and saponin aglycones are produced.

Detailed Description Text (12):
In this case, an enzyme known as .beta.-glucosidase, which decomposes the glycosides of isoflavone compounds and is manufactured by koji mold as a result of the propagation of koji mold in the soybean meal, breaks down the glycosides of the isoflavone compounds in the soybean meal and thus produces isoflavone aglycones. Furthermore, an enzyme known as .beta.-glucuronidase, which decomposes the glycosides of saponins and is manufactured by the koji mold, breaks down the glycosides of the saponins contained in the soybean meal and thus produces saponin aglycones.

Detailed Description Text (79):
As shown in Table 11 below, the measurement results were as follows: i.e., the amount of cytochrome P-450 was significantly lowest in the casein feed group. When the fermented defatted soybean group and the untreated defatted soybean group were compared, an increase in the amount of cytochrome P-450 was observed in the fermented defatted soybean group. It can be seen that this probably suggests that the amount of cytochrome P-450 in a dose-dependent manner with isoflavone aglycones. In other words, if the three test feeds are compared, it is seen that the fermented defatted soybeans manufactured by the process of the present invention cause the greatest increase (with a significant difference) in the amount of cytochrome P-450 (which is a hepatic drug metabolizing enzyme). Furthermore, cytochrome P-450 II also contributes to the promotion of metabolism in Practical Examples 3 and 4 described below; thus, it can be seen that fermented defatted soybean manufactured by the process of the present invention cause a significant increase in the promotion of metabolism.

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L1: Entry 4 of 18 File: USPT Jan 21, 2003

DOCUMENT-IDENTIFIER: US 6509043 B1
** See image for Certificate of Correction **
TITLE: Composition comprising soy protein, dietary fibres and a phytoestrogen compound and use thereof in the prevention and/or treatment of pulmonary diseases

Detailed Description Text (17):
Without wishing to be bound by any specific theory it is presently believed that both soluble dietary fibres (working as nutrients) and insoluble dietary fibres (working as bulking agents), in particular from soybean fibres, more particularly soy cotyledon fibres, provide favourable growth conditions for the microflora in the human gut, which makes the microflora more effective in deconjugating isoflavones in the glucoside form to the aglycone form. Isoflavones in the aglycone form are absorbed faster and to a greater extent in the human gut than isoflavones in the glucoside form, and isoflavones in the aglycone form are the biologically more active species. In view hereof it can be understood that administration of a combination of soy proteins, a high, fixed level of isoflavones and a combination of soluble and insoluble fibres is effective in providing an increased uptake of isoflavones.

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L1: Entry 5 of 18 File: USPT Dec 31, 2002

DOCUMENT-IDENTIFIER: US 6500965 B2
TITLE: Dry extract which is rich in isoflavones in the form of aglycones and process for preparing the same

Brief Summary Text (1):
The present invention relates to a dry extract which is rich in isoflavones, as aglycones, and to a process for preparing the same.

Brief Summary Text (2):
More particularly, the present invention relates to a dry extract containing at least 8% (w/w) of one or more isoflavones, as aglycones, of formula ##STR2##

Brief Summary Text (15):
However, the extraction yield is never indicated in this document. In addition, in the experimental section it is reported that the aqueous phase contains the isoflavones as aglycones (Example 1, page 12, line 26). However, this must be an entirely negligible amount since it is known that the aglycones are substantially insoluble in water.

Brief Summary Text (16):
There is therefore still a great need for a dry extract containing at least 8% (w/w) of one or more isoflavones, as aglycones, and for a process for preparing the same.

Detailed Description Text (2):
Preparation of a dry extract of Trifolium pratense (titre of isoflavones as aglycones=19.5 w/w %)

Detailed Description Text (7):
22.0 g of dry extract were thus obtained (yield=4.4% by weight relative to the weight of the starting dry Trifolium pratense) having a titre, by HPLC, of isoflavones as aglycones of 19.5% (w/w) [about 9% (w/w) of Biochanin, about 8% (w/w) of Formononetin, about 1% (w/w) of Daidzein and about 1.5% (w/w) of Genistein].

Detailed Description Text (8):
The yield of isoflavones as aglycones is thus about 0.86% (19.5%.times.4.4%) relative to the weight of the starting dry plant material.

Detailed Description Text (10):
Preparation of a dry extract of Trifolium pratense (titre of isoflavones as aglycones=21.6 w/w %)

Detailed Description Text (12):
17.5 g of dry extract were thus obtained (yield=3.5% by weight relative to the weight of the starting dry Trifolium pratense) with a titre, by HPLC, of isoflavones as aglycones of 21.6% (w/w) [about 10% (w/w) of Biochanin, about 9% (w/w) of Formononetin, about 1% (w/w) of Daidzein and about 1.5% (w/w) of Genistein].

Detailed Description Text (13):

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The yield of isoflavones as aglycones is thus about 0.76% (21.6%.times.3.5%) relative to the weight of the starting dry plant material.

Detailed Description Text (15):
Preparation of a dry extract of Trifolium pratense (titre of isoflavones as aglycones=38 w/w %)

Detailed Description Text (16):
The dry extract of Trifolium pratense (10 g) obtained as described in Example 1 (titre of isoflavones as aglycones of 19.5 w/w %) was treated with a mixture of 95% ethanol (200 ml), tap water (600 ml) and sodium chloride (0.5 g). The thus obtained mixture underwent extraction with methyl tert-butyl ether (3.times.400 ml). The combined ether extracts were washed with 20% ethanol (200 ml). The ether solution was separated out and concentrated under vacuum to give a dry residue. The dry residue was oven-dried under a residual vacuum of about 660 pascals.

Detailed Description Text (17):
5.0 g of dry extract were thus obtained (yield=50% by weight relative to the weight of the starting dry extract of Trifolium pratense and yield=2.2% by weight relative to the starting 500 g of Trifolium pratense from Example 1) having a titre, by HPLC, of isoflavones as aglycones of 38% (w/w) [about 18% (w/w) of Biochanin, about 16% (w/w) of Formononetin, about 1.5% (w/w) of Daidzein and about 2.5% (w/w) of Genistein].

Detailed Description Text (21):
6.0 g of dry extract were thus obtained (yield=60% by weight relative to the weight of the starting dry extract of Trifolium pratense and yield=2.64% by weight relative to the starting 500 g of Trifolium pratense from Example 1) having a titre, by HPLC, of isoflavones as aglycones of 32% (w/w) [about 15% (w/w) of Biochanin, about 13% (w/w) of Formononetin, about 2% (w/w) of Daidzein and about 2% (w/w) of Genistein].

Detailed Description Text (27):
14.02 g of dry residue were thus obtained (yield=28% by weight relative to the weight of the starting dry Trifolium pratense) having a titre, by HPLC, of isoflavones as aglycones of 0.4% (w/w) [about 0.2% (w/w) of Biochanin, about 0.18% (w/w) of Formononetin, about 0.01% (w/w) of Daidzein and about 0.01% (w/w) of Genistein].

Detailed Description Text (28):
The yield of isoflavones as aglycones is thus about 0.11% (28%.times.0.4%) relative to the weight of the starting dry plant material.

Detailed Description Text (34):
1.85 g of dry extract were thus obtained (yield=3.7% by weight relative to the weight of the starting dry Trifolium pratense) having a titre, by HPLC, of isoflavones as aglycones of 3.1 w/w% [about 1.6% (w/w) of Biochanin, about 1.4% (w/w) of Formononetin, about 0.05% (w/w) of Daidzein and about 0.05% (w/w) of Genistein].

Detailed Description Text (35):
The yield of isoflavones as aglycones is thus about 0.1147% (3.7%.times.3.1%) relative to the weight of the starting dry plant material.

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L1: Entry 6 of 18 File: USPT Dec 17, 2002

DOCUMENT-IDENTIFIER: US 6495140 B1
TITLE: Process for the isolation, recovery and purification of non-polar extractives

Detailed Description Text (40):
TLC of saponins was performed on MKC.sub.18 F reversed phase plates (1.times.3 in., 200 .mu.m thickness from Whatman International Ltd, Maidstone, England) developed with methanol: aqueous 5% acetic acid (75:25 v:v). Compounds were visualised by spraying with a 0.5% solution (v:v) of p-anisaldehyde in acidified aqueous ethanol (ethanol concentrated sulfuric acid:water:p-anisaldehyde 90:5:4.5:0.5 v:v:v:v) and heating at 100.degree. C. for 3 min. This reagent gives a number of distinct colors with different constituents including brown (free sugars), transitory yellow quickly turning to pink, green, or grayish-blue (saponins), slowly appearing reddish (amino acids, prolamines), purple (galactosylglycerides), and yellow (lysophosphatides). TLC of isoflavone aglycones and glycosides was carried out on 200 .mu.m thick silica gel plastic-backed plates (Baker-Flex 1B-2-F, VWR Scientific, Ottawa, Canada) using the following solvent systems: for aglycones, toluene:methyl ethyl ketone:acetic acid (80:15:5 v:v:v); for glycosides, dichloromethane:ethyl acetate:methanol:aqueous 5% acetic acid (40:35:20:5 v:v:v:v). Detection was made using a spray reagent consisting of 0.1% (w:v) diphenylborinic acid ethanolamine complex (Sigma Chemical Co., St. Louis, Mo.) in isopropanol and examining the air-dried plate in UV (365 nm) light.

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L1: Entry 7 of 18 File: USPT Sep 3, 2002

DOCUMENT-IDENTIFIER: US 6444239 B1
TITLE: Process for producing isoflavone aglycone-containing composition

Abstract Text (1):
An isoflavone aglycone-containing composition having genistein as a main aglycone is produced by a process comprising allowing a protease and .beta.-glucosidase to act on a soy protein raw material, an extract of a soy protein raw material or a by-product of a soy protein raw material to water-solubilize the protein of soybean origin and to convert isoflavone glycosides to the corresponding aglycones, separating water-soluble components from the enzymatic reaction mixture, and recovering water-insoluble matter.

Brief Summary Text (3):
The present invention relates to a process for obtaining a composition containing soybean isoflavone aglycones mainly comprising genistein.

Brief Summary Text (5):
Isoflavone compounds, such as malonyldaidzin, malonylglycitin, malonylgenistin, daidzin, glycitin, genistin, daidzein, glycitein, and genistein, are known to have estrogenic activity, antioxidative activity, antibacterial activity, antilipemia activity, anticholesterol activity, and the like. In recent years, cancer cell differentiation and induction activity, oncogene inhibitory activity, and prophylactic activity on cancers have been confirmed. Thus, the usefulness of these isoflavone compounds has been attracting attention. Many researches have revealed that the pharmacological effects, such a cancer prophylactic effect, of isoflavone compounds are primarily attributed not to the glycosides themselves but their aglycones, such as daidzein or genistein. Of the soybean isoflavone aglycones genistein has recently been proved particularly excellent in physiological activities, including antiosteoporosis activity, antiarteriosclerotic activity, and anticancer activities in the breast, the stomach and the prostate (see M. Numoto, Cancer Research, vol. 53, p. 5815 (1993) and S. Barnes, Biochem. Biophys. Res. Commun., vol. 179, p. 661 (1991).

Brief Summary Text (6):
Methods of obtaining isoflavone compounds include the methods described in Japanese Patent Laid-Open Nos. 62-126186 and 11-89589, for example. Since 95% or more of isoflavone compounds in soybeans are present in the form of glycosides, however, the isoflavone compounds obtained by the former method mainly comprise glycosides with little amount of aglycones. Further, the isoflavone aglycones obtained by the latter method, which uses soybean hypocotyl tissue as a raw material, mainly comprise daidzein with the genistein content being about 1/6 of daidzein.

Brief Summary Text (9):
The present inventors have conducted extensive investigations on production of isoflavone aglycones from soybeans and found as a result that a composition containing isoflavone aglycones the majority of which is genistein can easily be obtained by allowing an enzyme preparation having protease activity and .beta.-glucosidase activity to thoroughly act on a soybean raw material that is easily available in the market and recovering water-insoluble matter.

Brief Summary Text (10):

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Having been completed based on the above finding, the present invention provides a process for producing an isoflavone aglycone-containing composition comprising allowing a protease and .beta.-glucosidase to act on a soy protein raw material, an extract of a soy protein raw material or a by-product of a soy protein raw material to water-solubilize the protein of soybean origin and to convert isoflavone glycosides to the corresponding aglycones, separating water-soluble components from the enzymatic reaction mixture, and recovering water-insoluble matter. The majority of the aglycones present in the resulting composition is genistein.

Brief Summary Text (11):
According to the present invention, a composition containing isoflavone aglycones mainly comprising genistein can be obtained from an easily commercially available raw material through an easy and simple operation.

Brief Summary Text (20):
The enzymatic reaction can be regarded completed when a sampled aliquot of the reaction mixture shows conversion of 90% or more of the isoflavone glycosides into the corresponding aglycones. On completion of the reaction, the reaction mixture is adjusted to pH 2 to 5, and water-soluble components are removed by ultrafiltration, centrifugal separation or a like technique to recover water-insoluble matter. If desired, the recovered water-insoluble matter is washed with water at pH 2 to 5. The separated water-soluble components (the soluble matter of the reaction mixture plus the washing) include water-solubilized amino acids and peptides resulting from decomposition by the protease and the sugar moieties resulting from hydrolysis by the .beta.-glucosidase. Accordingly, the above operation removes components other than isoflavone aglycones and provides the insoluble matter enriched in aglycones.

Brief Summary Text (25):
After confirming that 90% or more of the isoflavone glycosides in the reaction mixture has converted to the corresponding aglycones, the reaction mixture is adjusted to pH 2 to 5, and water-soluble components are removed by ultrafiltration, centrifugal separation or a like technique to recover water-insoluble matter. If desired, the recovered water-insoluble matter is washed with water at pH 2 to 5. The resulting wet solid is dried by means of, for example, a vacuum drier and pulverized to obtain an isoflavone aglycone-containing composition. Rich in isoflavone aglycones, the resulting powder can be utilized as health foods and general beverages and foodstuffs. The resulting powder or an intermediate product obtained in the course of the above-mentioned preparation process may be purified by organic solvent extraction or by use of a resin to provide high purity isoflavone aglycone preparations, which can be used as health foods, cosmetics or ingredients of pharmaceutical preparations.

Detailed Description Text (2):
Defatted soybeans (200 g) were ground and extracted with 1000 ml of 80% ethanol to obtain 23 g of an extract containing isoflavone glycosides. To the extract was added 500 ml of water to dissolve the extract. The solution was adjusted to pH 4.5, 2 g of Sumizyme FP (available from Shin-nihon Kagaku Kogyo K.K.) was added thereto, and the mixture was stirred at 55.degree. C. overnight. It was found that 90% or more of the isoflavone glycosides had been converted to aglycones. The reaction mixture was adjusted to pH 4 with hydrochloric acid to precipitate an isoflavone fraction, which was collected by filtration. The filter cake was dissolved in 1000 ml of 0.1N NAOH, adjusted to pH 8, and passed through an activated carbon column. After washing the column with water, the adsorbed isoflavone aglycones were eluted with 2000 ml of 0.1N NaOH. The eluate was adjusted to pH 4.5 with hydrochloric acid to precipitate the aglycones, which were collected by filtration and dried in a vacuum drier to give 300 mg of an isoflavone aglycone powder having a purity of 70%. The isoflavone aglycone content of the resulting powder was found made up of 58% of genistein, 38% of daidzein, and 4% of glycitein, proving to be a composition mainly comprising genirstein. The analysis of the aglycone composition was in accordance with H. Wang et a l., J. Agric. Food Chem., vol. 42, p. 666 (1994).

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Detailed Description Text (4):
Three liters of water was added to 100 g of a commercially available soy protein isolate Fuji Pro F (available from Fuji Oil Co., Ltd.), and the mixture was heated in a boiling water bath for 10 minutes. After cooling, 10 g of Molsin F (available from Kikkoman Corp.) and 300 ml of ethanol were added thereto at a temperature kept at 50.degree. C., followed by stirring at pH 5 overnight (16 hours). After the reaction, 95% or more of the isoflavones in the reaction mixture were found to have been converted to the corresponding aglycones. The reaction mixture was adjusted to a pH of 4.5. The precipitate was collected by centrifugation, washed with water at pH 4.5, and dried in a freeze-drier to obtain a product weighing 34 g. The product was found to have an isoflavone aglycone content of about 0.61%, which is about three times that of the raw material. Analysis by high-performance liquid chromatography revealed that the isoflavone aglycone composition of the product was made up of 56% of genistein, 39% of daidzein, and 5% of glycitein, proving to comprise genistein as a main aglycone.

Detailed Description Text (6):
To 1 kg of a commercially available isoflavone glycoside Novasoy (available from Archer Daniels Midland Co.) was added 25 l of distilled water. After the mixture was adjusted to pH 4.5 with hydrochloric acid, 150 g of Molsin F (Kikkoman Corp.) was added thereto, followed by stirring at 60.degree. C. overnight (16 hours). After the reaction, 95% or more of the isoflavones in the reaction mixture were found converted to aglycones. The reaction mixture was adjusted to a pH of 2.5 with hydrochloric acid to precipitate an isoflavone fraction, which was washed with water at pH 2.5 and collected by filtration. The filter cake was stirred in 25 l of 90% ethanol for 2 hours to extract the isoflavones. The insoluble enzyme component was removed, the separated ethanol extract was concentrated, and the concentrate was spray-dried to obtain 480 g of an isoflavone aglycone powder. The resulting powder had an isoflavone aglycone content of 52%, which was found to be made up of 54% of genistein, 40% of daidzein, and 6% of glycitein, proving to comprise genistein as a main aglycone.

CLAIMS:

1. A process for producing an isoflavone aglycone-containing composition comprising allowing a protease and .beta.-glucosidase to act on a soy protein raw material, an extract of a soy protein raw material or a by-product of a soy protein raw material to water-solubilize the protein of soybean origin and to convert isoflavone glycosides to the corresponding aglycones in an enzymatic reaction mixture, separating water-soluble components from the enzymatic reaction mixture, and recovering water-soluble matter comprising isoflavone aglycones, wherein a majority of said isoflavone aglycones is genistein.

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L1: Entry 8 of 18 File: USPT Jun 25, 2002

DOCUMENT-IDENTIFIER: US 6410699 B1
TITLE: Process for the preparation of isoflavone compounds

Brief Summary Text (3):
The present invention relates to a process for preparation of isoflavone compounds from materials that contain isoflavone compounds, especially from those plants that are members of such families as Leguminosae, Rosaceae, Iridaceae, Morus, and Amarantus inamoenus. More particularly, the present invention relates to a-process for preparation of, isoflavone compounds, from leguminous plants, especially isoflavone aglycones that are hydrolysates of isoflavone glycosides.

Brief Summary Text (10):
Among the isoflavone compounds having the above formula (II), those ones whose R, is glucose are referred to as isoflavone glycosides, and hydrolysates of isoflavone glycosides are referred to as isoflavone aglycones.

Brief Summary Text (12):
There are many reports on the pharmacological effects of isoflavone compounds, especially isoflavone aglycones that are hydrolysates of isoflavone glycosides.

Brief Summary Text (17):
Since isoflavone compounds have pharmacological effects as mentioned above, there is now an increasing demand for the supply of isoflavone compounds in the medicine and food industries. However, isoflavone compounds are present in natural substances only in small quantities; in particular, isoflavone aglycones are contained in natural substances in extremely small quantities.

Brief Summary Text (18):
For instance, 95% or more of isoflavone compounds present in soybeans, which belong to Leguminosae, are isoflavone glycosides, and isoflavone aglycones are only 5% or less of the isoflavone compounds. For this reason, there is now a demand for a simple process for efficiently recovering isoflavone compounds, especially isoflavone aglycones, from materials containing isoflavone compounds in effective amounts.

Brief Summary Text (21):
It is possible to obtain isoflavone compounds by the process described in Japanese Patent Laid-Open Publication No. 126186/1987. However, most of the isoflavone compounds obtainable by this method are daidzin and genistin, and isoflavone aglycones, which are isoflavone compounds desired, can be obtained only in low recovery percentages.

Brief Summary Text (23):
This report describes that, during fermentation of soybeans to produce soy sauce or miso, isoflavone glycosides are fully hydrolyzed to form isoflavone aglycones. However, this report teaches only a method for qualitatively or quantitatively analyzing isoflavone compounds to show the progress of the hydrolysis of isoflavone glycosides to isoflavone aglycones in each step of production of soy sauce or the like. This method is not suitable at all for the industrial production of isoflavone aglycones.

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Brief Summary Text (24):
Japanese Patent Laid-Open Publication No. 170756/1993 describes a method for extracting isoflavone aglycones from isoflavone compounds contained in soy sauce cake or soy sauce oil. To obtain isoflavone aglycones in increased recovery percentages, the fact described in the above-described report that isoflavone glycosides are hydrolyzed to form isoflavone aglycones in the course of the production of soy sauce is applied to this method. However, the recovery percentages of isoflavone aglycones attainable by this method are not yet satisfactorily high.

Brief Summary Text (25):
Japanese Patent Laid-Open Publication No. 258669/1989 discloses a process for producing and recovering isoflavone aglycones, in which isoflavone glycosides are hydrolyzed with the aid of .beta.-glucosidase, one of enzymes which soybeans themselves have. Practically, however, the yields of the isoflavone aglycones produced by this process are low.

Brief Summary Text (26):
The specifications of Japanese Patent Applications No. 32385/1994, No. 179111/1995, No. 26888/1995, No. 88552/1996, No. 83036/1997, etc. which the applicant of the present invention previously filed with the Japanese Patent Office disclose processes for producing concentrated isoflavone compounds containing isoflavone aglycones in large quantities. In these processes, isoflavone glycosides, which are present in legume in large quantities, are hydrolyzed by inoculating grains (soybeans, etc.) with microorganisms such as koji-kin, a fungus belonging to the genus Aspergillus oryzae. These are improved or modified processes established on the basis of the fact that isoflavone aglycones are efficiently obtainable from isoflavone glycosides.

Brief Summary Text (27):
Undoubtedly, the aforementioned prior techniques are improved methods useful for separating and recovering isoflavone compounds, especially isoflavone aglycones, from starting materials (methods for increasing the content of isoflavone aglycones by subjecting isoflavone glycosides to hydrolysis by various means being included). However, as far as we know, isoflavone aglycones cannot be obtained in satisfactorily high recovery percentages even by these conventional methods.

Brief Summary Text (29):
This must be the reason why the recovery of isoflavone glycosides, especially isoflavone aglycones that are hydrolysates of isoflavone glycosides, has not been improved.

Brief Summary Text (31):
We paid our attention to a method in which fat is, first of all, fully removed from a starting material, and the defatted material is, in the next step, subjected to solvent extraction to prepare isoflavone compounds in high recovery percentages, and finally accomplished the present invention. The present invention is to provide a process for preparation of especially isoflavone aglycones in high recovery percentages. By also taking the utilization of isoflavone aglycones in food products, medicines, etc. into consideration, safety solvents are used in the process of the present invention.

Brief Summary Text (40):
In the process for preparation of isoflavone compounds according to the present invention, especially in the process for producing isoflavone aglycones by the hydrolysis of isoflavone glycosides, fat is removed from the starting material before separating therefrom isoflavone compounds by means of extraction. By doing so, it is possible to successfully solve various problems in the prior art.

Brief Summary Text (41):

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It has now been found the following: like in the case of the process in which a starting material containing isoflavone compounds is firstly subjected to extraction with an alcohol (see Japanese Patent Laid-Open Publication No. 170756/1993), when a starting material containing isoflavone compounds (e.g., soy sauce cake) is directly subjected to extraction with a polar solvent, lipids contained in the starting material are also extracted; therefore, even if fat is removed, after the extraction, from the extract by the use of a non-polar solvent (e.g., n-hexane), it is not easy to separate lipids from the isoflavone compounds, and this makes the recovery percentages of isoflavone compounds, especially isoflavone aglycones not so high.

Brief Summary Text (42):
The reason for this is as follows: since isoflavone aglycones are less polar than isoflavone glycos ides for instance, in a thin layer chromatographic analysis of a sample of isoflavone compounds including both isoflavone glycosides and isoflavone aglycones, the change in polarity can well be confirmed if a plurality of liposoluble components are present between the spots of the two compounds), isoflavone aglycones tend to intermingle with liposoluble components, making it difficult to separate isoflavone aglycones from lipids.

Brief Summary Text (43):
On the contrary, the process for preparation of isoflavone compounds according to the present invention is characterized in that fat is, at first, fully removed from a starting material as mentioned above. The process of the present invention is therefore almost free from the above-described problem, and can give the desired substances, isoflavone aglycones, in high recovery percentages.

Brief Summary Text (57):
Those soybeans that have been hydrolyzed to contain isoflavone aglycones in significant amounts are particularly preferred, and typically preferred are soy sauce oil, soy sauce cake, tamari cake, miso, mame-miso, natto, fermented soybeans (including soybeans fermented by microorganisms such as koji-kin, a fungus belonging to the genus Aspergillus oryzae), and the like.

Brief Summary Text (64):
A solvent typically used for this solvent extraction method is a non-polar solvent. Any non-polar solvent can be used as long as it can fully extract unnecessary soybean oil and other lipids contained in the starting material, and as long as it has polarity low enough to avoid the solubilization of isoflavone aglycones.

Brief Summary Text (66):
Particularly preferred in the present invention is n-hexane. n-Hexane does not solubilize isoflavone aglycones.

Brief Summary Text (76):
A polar solvent is typically used for this solvent extraction, and any polar solvent can be used as long as it can extract isoflavone compounds, especially isoflavone aglycones, present in the dried defatted material.

Brief Summary Text (78):
Ethyl acetate is a polar solvent particularly suitable for extracting isoflavone aglycones. In addition, it has a low boiling point; and it has been used as an extraction solvent in the food industry, and is thus highly safe.

Brief Summary Text (94):
After dissolving, in the polar solvent, the extract concentrated to dryness, the solution obtained is diluted with water to precipitate, as insoluble matter, isoflavone aglycones whose polarity is slightly lower than that of isoflavone glycosides.

Brief Summary Text (98):
A conventional method is used to separate, from the above-obtained dilute solution, the insoluble matter containing considerable amounts of isoflavone compounds whose isoflavone aglycones content is high.

Detailed Description Text (16):
The data in Table B show that the contents of the individual isoflavone compounds in the concentrated isoflavone compounds obtained in accordance with the process of the invention are unusually higher than those of the individual isoflavone compounds in the fermented soybean hypocotyl, starting material. Specifically, the contents of the isoflavone glycosides in the former are as high as about 10 to 13 times the contents of the same in the latter, and the contents of the isoflavone aglycones in the former are about 57 to 64 times the contents of the same in the latter. These results thus demonstrate that isoflavone compounds can simply and efficiently be obtained by the process of the present invention in high recovery percentages.

Detailed Description Text (28):
As can be understood from the above examples, in the process for preparation of isoflavone compounds according to the present invention, fat is, at first, fully removed from a starting material containing isoflavone compounds and/or their precursors in significant amounts, and the defatted material is then subjected to separation of isoflavone compounds by means of extraction, so that isoflavone compounds, especially isoflavone aglycones, can be obtained in higher recovery percentages than those in prior art processes, as previously mentioned in THE SUMMARY OF THE INVENTION, EXAMPLES, and the like.

Detailed Description Text (29):
Thus, by the use of the process of the present invention, highly concentrated isoflavone compounds, especially isoflavone aglycones, which are excellent in carcinostatic effect, therapeutic effect for osteoporosis, immunosuppressive effect and other medical effects, can be obtained easily and inexpensively from starting materials containing isoflavone compounds and/or their precursors in significant amounts. The isoflavone compounds obtained by the process of the present invention can be supplied in; large quantities as starting materials to the medicine and food industries, in which the supply of isoflavone compounds is now strongly demanded. Further, the isoflavone compounds obtained by the process of the present invention can be used as starting materials for producing individual isoflavone aglycones.

CLAIMS:

1. A process for preparation of an isoflavone aglycone compound, comprising the steps of:

(1) removing fat from fermented soybeans to form a defatted material and drying the defatted material;

(2) subjecting the defatted material obtained in step (1) to extraction with a solvent to form an extract containing one or more isoflavone compounds and concentrating the extract to dryness;

(3) dissolving the extract from in step (2) in a second solvent to form a solution, diluting the solution with water, and separating insoluble matter that precipitates from the solution; and

(4) washing the insoluble matter obtained in step (3), and drying it to remove the second solvent, thereby obtaining one or more isoflavone aglycone compounds.

3. A process for preparation of an isoflavone aglycone compound, comprising the steps of:

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(1) removing fat from fermented soybeans with a non-polar solvent to form a defatted material and drying the defatted material;

(2) subjecting the defatted material obtained in step (1) to extraction with a polar solvent to form an extract containing one or more isoflavone compounds and concentrating the extract to dryness;

(3) dissolving, the extract from step (2) in a second polar solvent to form a solution, diluting the solution with water, and separating insoluble matter that precipitates from the solution; and

(4) washing the insoluble matter obtained in step (3), and drying it to remove the second polar solvent, thereby obtaining one or more isoflavone aglycone compounds.

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L1: Entry 9 of 18 File: USPT Jan 22, 2002

DOCUMENT-IDENTIFIER: US 6340703 B1
TITLE: Treatment or prevention of osteoporosis

CLAIMS:

12. A method according to claim 6, wherein the isoflavones are in the aglycone, glycoside, malonyl or acetyl form.

13. A composition according to claim 9, wherein the isoflavones are in the aglycone, glycoside, malonyl or acetyl form.

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L1: Entry 10 of 18 File: USPT Dec 4, 2001

DOCUMENT-IDENTIFIER: US 6326366 B1
TITLE: Hormone replacement therapy

Detailed Description Text (23):
After conversion of the isoflavone glycoside conjugates and isoflavone glycosides to their respective aglycone isoflavones, the aglycone isoflavones can be extracted from the plant material as described above. The water in the plant material slurry may be evaporated prior to extracting the plant material with an extractant, or the water may be utilized together with another solvent as the extractant.

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L1: Entry 11 of 18 File: USPT Oct 16, 2001

DOCUMENT-IDENTIFIER: US 6303161 B1
** See image for Certificate of Correction **
TITLE: Product containing healthful component and process for preparing the same

Brief Summary Text (44):
In concrete terms, proteins, isoflavones, saponins and phytic acid, etc., are respectively converted into the health-promoting component such as peptides, isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc., by performing a koji preparation treatment and a hydrolysis treatment on beans used as a raw material, and are thus converted into a state in which these components can be very easily absorbed by the digestive tracts of single-stomached animals, etc. Using the product of the present invention which has such a health-promoting component based on the above-described koji preparation treatment and hydrolysis treatment, health promotion by means of at least one member of the group consisting of liver function improving constituents, cardiac function improving constituents, anti-inflammatory constituents, antifat constituents, antioxidation consititents and antimutagen constituents can be accomplished very effectively.

Brief Summary Text (49):
Since the product and preparation process of the present invention are constructed and act as described above, a health-promoting component can be produced in a product using beans as a raw material, by means of a koji preparation treatment and a hydrolysis treatment. In concrete terms, peptides, isoflavones, saponins and phytic acid, etc. are respectively converted into a health-promoting component such as isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc. and are thus converted into a state which can be very easily absorbed by the digestive tracts of single-stomached animals. In the present invention, furthermore, the intestine-regulating bacteria added following the initiation of the above-described koji preparation treatment but prior to the completion of the hydrolysis treatment propagate so that a health-promoting component consisting of such intestine-regulating bacteria can be included in the product in large amounts, thus making it possible to achieve an intestine-regulating effect in the intestines of single-stomached animals. By using the product of the present invention which contains such a health-promoting component, health promotion by means of at least one member of the group consisting of liver function improving constituents, cardiac function improving constituents, anti-inflammatory constituents, antifat constituents, antioxidation consititents and antimutagen constituents and components which have an intestine-regulating effect in single-stomached animals can be accomplished very effectively. This product can be used directly "as is", or the efficacy can be strengthened by extracting and concentrating the isoflavone aglycones, saponin aglycones, myo-inositol or Maillard reaction products, etc; in addition, this product can also be utilized in applied products which use this product as a raw material, e. g., food products, livestock feeds, pet foods or drug raw materials, etc. and is thus superior in terms of all-purpose utility. Furthermore, miso and soy sauce are foods with a high salt content, since salt is added following the koji preparation step, so that such food products lack all-purpose utility; moreover, the aging period of such food products following the koji preparation step is extremely long, so that there are problems in productivity. However, the product of the present invention is easy to manufacture at a low-cost.

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Detailed Description Text (9):
Afterward, the mixture is placed in a koji preparation device and held for a prescribed period of time with the initial temperature set at approximately 28 to 30.degree. C. Koji preparation is then performed by fermenting the soybean meal with a low moisture content of 40 wt % by means of koji mold until the enzymes required in order to convert the proteins, isoflavones, saponins and phytic acid, etc., contained in the soybean meal into respective health-promoting components such as peptide, isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc., are produced.

Detailed Description Text (11):
With respect to such cases in which a health-promoting component is produced by this koji preparation step, a case will be described in which the glycosides of isoflavones and saponins are decomposed so that respective isoflavone aglycones and saponin aglycones are produced.

Detailed Description Text (12):
In this case, an enzyme known as .beta.-glucosidase, which decomposes the glycosides of isoflavone compounds and is manufactured by koji mold as a result of the propagation of koji mold in the soybean meal, breaks down the glycosides of the isoflavone compounds in the soybean meal and thus produces isoflavone aglycones. Furthermore, an enzyme known as .beta.-glucuronidase, which decomposes the glycosides of saponins and is manufactured by the koji mold, breaks down the glycosides of the saponins contained in the soybean meal and thus produces saponin aglycones.

Detailed Description Text (51):
Furthermore, a health-promoting component such as peptides, isoflavone aglycones, saponin aglycones, myo-inositol and Maillard reaction products, etc., as well as a health-promoting component consisting of lactic acid bacteria, which are one type of intestine-regulating bacteria, and bacteriocin, which is a product of such bacteria, are contained in such food products.

Detailed Description Text (79):
As shown in Table 11 below, the measurement results were as follows: i. e., the amount of cytochrome P-450 was significantly lowest in the casein feed group. When the fermented defatted soybean group and the untreated defatted soybean group were compared, an increase in the amount of cytochrome P-450 was observed in the fermented defatted soybean group. It can be seen that this probably suggests that the amount of cytochrome P-450 in a dose-dependent manner with isoflavone aglycones. In other words, if the three test feeds are compared, it is seen that the fermented defatted soybeans manufactured by the process of the present invention cause the greatest increase (with a significant difference) in the amount of cytochrome P-450 (which is a hepatic drug metabolizing enzyme). Furthermore, cytochrome P-450 II also contributes to the promotion of metabolism in Practical Examples 3 and 4 described below; thus, it can be seen that fermented defatted soybean manufactured by the process of the present invention cause a significant increase in the promotion of metabolism.

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L1: Entry 12 of 18 File: USPT Aug 28, 2001

DOCUMENT-IDENTIFIER: US 6280777 B1
**** See image for Certificate of Correction ****
TITLE: Soya extract, process for its preparation and pharmaceutical composition

Brief Summary Text (6):
According to biomedical literature and epidemiological information published in recent years, principally in relation to populations of the East, which consume soya-based foods to a great extent, the use of these foods to a high degree reduces pre-menopausal and post-menopausal symptoms in women (A. Cassidy, Proceedings of the Nutrition Society, 1996, 55, 339-417) These facts, which still lack a clear scientific basis, are usually ascribed to the isoflavone aglycones genistein, daidzein and glycitein, which are present in the various soya-based foods.

Brief Summary Text (8):
According to further biomedical literature and epidemiological information published in recent years, principally relating to population groups in the East, which consume soya-based foods to a great extent, the use of these foods decreases to a high degree breast cancer in women and cancer of the prostate in men (A. Nomura, B. E., Henderson J. Lee, American Journal of Clinical Nutrition, 1978, 31, 2020-2025; T. Eirayama in Diet, Nutrition and Cancer, 1986 pp. 41-53, Y. Hayashi, M. Nagao, T. Sugimura, S. Takayama, L. Tomatis, L. W. Wattenberg and G. N. Wogan eds. Tokyo: Japanese Scientific Society Press; R. K. Severson, A. M. Y. Nomura, J. S. Grove, G. N. Stemmerman, Cancer Research, 1989, 49, 1857-1860). Also, these facts, which still lack a clear scientific basis, are usually ascribed to the isoflavone aglycones genistein, daidzein and glycitein which are present in the various soya-based foods.

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L1: Entry 13 of 18 File: USPT Apr 4, 2000

DOCUMENT-IDENTIFIER: US 6045819 A
TITLE: Substance containing health-promoting component and process for the production thereof

Detailed Description Text (20):
Of the formation of the healthful constituents, description will be given with respect to the formation of the aglycones of isoflavones and the aglycones of soyasaponins by hidrolytically separating glycosidic saccharides from isoflavones and saponins, respectively.

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L1: Entry 14 of 18 File: USPT Jan 25, 2000

DOCUMENT-IDENTIFIER: US 6017893 A
TITLE: Use of isoflavones to prevent hair loss and preserve the integrity of existing hair

CLAIMS:

6. The method of claim 1, wherein said isoflavone is an aglycone.

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L1: Entry 15 of 18 File: USPT Mar 23, 1999

DOCUMENT-IDENTIFIER: US 5885632 A
TITLE: Process for preparing a product from a pulse crop as a starting material and a food containing the product prepared from a pulse crop as a starting material

Brief Summary Text (20):
Although ingestion of foods in a satisfactory amount which contain a sufficient amount of such isoflavone aglycones having excellent pharmacological activities as mentioned above enables dietarily desired life to be realized which exhibits excellent effect in terms of health maintenance of a human being, no food has heretofore satisfied this expectation.

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L1: Entry 16 of 18 File: USPT Aug 4, 1998

DOCUMENT-IDENTIFIER: US 5789581 A
TITLE: Process for obtaining malonyl isoflavone glycosides and obtaining isoflavone glycosides or isoflavone aglycons from malonyl isoflavone glycosides

Brief Summary Text (3):
The present invention relates to a process for obtaining malonylisoflavone glycosides from an aqueous extract of soybean, and to a process for obtaining isoflavone glycosides as well as isoflavone aglycones from the malonylisoflavone glycosides.

Brief Summary Text (6):
Furthermore, it has been recently confirmed that malonylisoflavone glycosides such as malonyldaidzin and malonylgenistin represented by the formulae: ##STR1## are present in soybean, and it has been proved that these glycoside derivatives comprise the main components of isoflavone compounds in soybean. These malonylisoflavone glycosides are easily soluble in water and exhibit per se anti-oxidizing activity, so that they are anticipated to have pharmacological effects described above from the structural similarity to the afore-mentioned isoflavone and aglycone compounds.

Brief Summary Text (9):
In addition, the malonylisoflavone glycosides can be easily treated with an alkali or by heating to cut the ester linkage between malonyl moiety and glucopyranosyl moiety and to form isoflavone glycosides such as daidzin and genistin, which are further treated with an acid or an enzyme to form isoflavone aglycones. In other words, the malonylisoflavone glycosides can also be used as the raw materials for obtaining daidzin or genistin.

Brief Summary Text (13):
In consideration of the current situations, the present inventors have studied on the processes for obtaining a malonylisoflavone glycoside from soybean in a simple operation. As a result, they have found that the extraction of soybean with water makes possible of the selective extraction of the malonylisoflavone glycoside relatively easily, that when the aqueous extract is directly put into contact with an adsorbent, the malonylisoflavone glycoside in the extract is easily adsorbed on the adsorbent, which is then rinsed with an aqueous alcohol solution to elute the malonylisoflavone glycoside in efficiency, and that the malonylisoflavone glycoside may be treated for example with an alkali to convert it easily into an isoflavone glycoside or an isoflavone aglycone. The present invention has thus been accomplished.

Detailed Description Text (18):
According to the present invention, the malonylisoflavone glycoside, or malonyldaidzin and malonylgenistin can be obtained fractionatingly from the extract of soybean in a simple procedure, and an isoflavone glycoside as well as an isoflavone aglycone can also be obtained starting from the above described compounds by the alkali treatment or the like.

Detailed Description Text (21):
The isoflavone glycoside thus obtained can be converted into an isoflavone aglycone by the further treatment with an acid or an enzyme.

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Detailed Description Text (24):
In the case of the enzyme treatment, the isoflavone glycoside is dispersed in a solution of .beta.-glucosidase derived from soybean in 1/10M phosphate buffer (pH 5.0). After reaction at 50.degree. C. for about 6 hours, the reaction mixture is adsorbed on an ODS resin column, eluted with an aqueous alcohol solution, concentrated under reduced pressure, and lyophilized to give an isoflavone aglycone as a purified product.

Detailed Description Text (55):
The isoflavone glycosides and the isoflavone aglycones obtained in Examples 6-8 have the same physical properties as those of the authentic samples (available from FUNAKOSHI K.K.).

Detailed Description Text (56):
According to the present invention, it is possible to obtain efficiently in a simple procedure from an aqueous extract of soybean the malonylisoflavone glycosides, from which the isoflavone glycosides as well as isoflavone aglycones are also obtained by treatments with for example an alkali and the like.

CLAIMS:

5. A process for obtaining an isoflavone aglycone comprising treating a malonylisoflavone glycoside obtained according to the process of claim 1 with heat and/or an alkali to give an isoflavone glycoside, which is treated with an acid or an enzyme to give an isoflavone aglycone.

6. A process according to claim 5, wherein an isoflavone glycoside solution is treated with heat in an aqueous alcohol solution containing dilute hydrochloric acid to give an isoflavone aglycone.

7. A process according to claim 5, wherein an isoflavone glycoside solution is hydrolyzed with .beta.-glucosidase to give an isoflavone aglycone.

8. A process for obtaining an isoflavone aglycone, wherein a malonylisoflavone glycoside solution obtained according to the process of claim 1 is treated with heat directly in an aqueous alcohol solution containing dilute hydrochloric acid to give an isoflavone aglycone.

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L1: Entry 17 of 18 File: USPT Oct 21, 1997

DOCUMENT-IDENTIFIER: US 5679806 A
TITLE: Process for the isolation and purification of isoflavones

Detailed Description Text (6):
There is some evidence in the research literature that the isoflavone aglycones may have more biological activity than their respective glycosides. However, isoflavone aglycones are found in very low abundance in plant tissues. Therefore, the present invention also contemplates the hydrolysis of the isoflavone glycosides to form their respective aglycones. Cleavage of the glucose, acetylglucose or malonylglucose molecule from an isoflavone glycoside may be accomplished by subjecting the isoflavone glycoside to acid hydrolysis in HCl-water, preferably 4N HCl at 100.degree. C. for 5 hours. The benefits of an acid/water hydrolysis are the elimination of organic solvents and ease of recovery of the product by filtration. Following the hydrolysis, the isoflavone aglycone is recovered by filtration, dried, and may then be crystallized from methanol, as discussed above, or alternatively extracted with diethyl ether, evaporated, and recrystallized from alcohol as discussed in further detail in the Examples which follow.

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L1: Entry 18 of 18 File: USPT Aug 20, 1996

DOCUMENT-IDENTIFIER: US 5547671 A
TITLE: Anti-intoxication composition

Brief Summary Text (7):
Keung and Vallee, in an article in Proc. Natl. Acad. Sci. USA, Vol. 90, pp. 10008-10012, November 1993, Biochemistry, have reported experiments they conducted with Syrian Golden hamsters, ascribing alcohol-suppressant effects of such herbal compositions to daidzin and daidzein, respectively a glycosylated isoflavone and a aglycone thereof. But they do not know whether these substances per se are the pharmacologically active molecules which directly suppress ethanol intake or whether they act as prodrugs converted in vivo to pharmacologically active species.

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L15: Entry 1 of 1 File: USPT Mar 23, 1999

DOCUMENT-IDENTIFIER: US 5885632 A
TITLE: Process for preparing a product from a pulse crop as a starting material and a food containing the product prepared from a pulse crop as a starting material

Abstract Text (1):
A process for preparing a product from a pulse crop as a starting material and a food containing the product prepared from a pulse crop as a starting material are disclosed according to the present invention, thereby enabling a food, a livestock feed, an aquacultural feed or the like to be efficiently prepared, which is made from a leguminous crop or a defatted product thereof or the like, which has excellent carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect and immunosuppressive effect, and which can be ingested in a sufficient amount, and thereby enabling a wholesome food such as a biscuit having the above-mentioned excellent pharmacological activities. Any conventional product made from a pulse crop does not have such excellent pharmacological activities and a process for preparing the same is poor in efficiency. Significant characteristic feature of the present invention resides in that glycosidic saccharides are hydrolytically separated from isoflavone compounds contained in a pulse crop to form isoflavone compounds containing aglycones in a large amount and that phytic acid contained in a pulse crop is removed to obtain a product having excellent pharmacological activities.

Brief Summary Text (3):
The present invention relates to a process for preparing a product from a pulse crop as a starting material and a food containing the product prepared from a pulse crop as a starting material.

Brief Summary Text (4):
In the present invention, the term "pulse crop" means leguminous crops such as soybean, defatted products thereof and the like, and the term "product made from a pulse crop as a starting material" means foods, livestock feeds, aquacultural feeds and the like which are made from the above-mentioned pulse crop.

Brief Summary Text (6):
In general, soybean which is one of the pulse crops contains isoflavone compounds including daidzin, daidzein, genistin and genistein.

Brief Summary Text (11):
According to this report, however, although hydrolysis of a glycosidic saccharide proceeds to some extent by cooking of a defatted soybean or in a koji preparation step, most of the saccharide has already hydrolytically been separated in soy sauce sediment or soybean miso. Accordingly, it is difficult to employ these for a process for preparing a product from a pulse crop as a starting material.

Brief Summary Text (12):
Further, many reports have been made on pharmacological activities of aglycones derived from hydrolysis of glycosidic saccharides from isoflavone compounds.

Brief Summary Text (22):
With respect to osteoprosis, it is desired to remove phytic acid, which inhibits

calcium from being absorbed in a body, from a pulse crop.

Brief Summary Text (23):
In soybean which is one of beans, phytic acid is contained in an amount of about 1 to 2% by weight. Phytic acid is residually present also in a product made from soybean and inhibits activities of a vitamin B complex contained in the product to prevent absorption of minerals and the like contained in the product. Further explanatively, phytic acid is a compound having such a structure that myo-inositol has its all hydroxyl groups each bonded with a phosphoric acid group, and chelates with a nutritionally important trace metal element to form hardly soluble compound. Accordingly, when a food with high phytic acid content is ingested by a human being or animal, normal intestinal absorption of such metals, for example, calcium, magnesium, iron, zinc and the like is prevented to cause various deficiencies. It has further been found that phytic acid present in a product including a soy protein isolate prevents a monogasteric animal from utilizing zinc in a food. Further, phytic acid is known to have inhibitory activities on various digestive enzymes in a gastrointestinal digestive tract on which ions of minerals such as calcium act as activators and which include .alpha.-amylase, pepsin and trypsin. It is, therefore, desired to remove phytic acid from the product.

Brief Summary Text (25):
The present invention has been made in view of these points. It is, therefore, an object of the present invention to provide a process for preparing a product from a pulse crop such as a food, a livestock feed, an aquacultural feed or the like, which is made from a pulse crop, which has excellent carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect and immunosuppressive effect, and which can be ingested in a sufficient amount.

Brief Summary Text (26):
It is another object of the present invention to provide a healthful food, such as a biscuit or the like which contains a product that is made from a pulse crop and that has properties excellent in carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect and immunosuppressive effect.

Brief Summary Text (27):
To attain the above object, the process of the present invention for preparing a product from a pulse crop as a starting material comprises:

Brief Summary Text (28):
inoculating a koji starter on a pulse crop to prepare koji; and

Brief Summary Text (29):
adding water to the resultant from the koji preparation treatment to advance hydrolysis of a protein contained in the resultant;

Brief Summary Text (30):
wherein in the course of the koji preparation and the proteolysis, separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop is advanced to form isoflavone compounds containing aglycones in a large amount, thereby obtaining a product from the pulse crop as a starting material.

Brief Summary Text (31):
Further, the process of the present invention for preparing a product from a pulse crop as a starting material comprises:

Brief Summary Text (32):
inoculating a koji starter on a pulse crop to prepare koji; and

Brief Summary Text (33):
adding water to the resultant from the koji preparation treatment to advance

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hydrolysis of a protein contained in the resultant;

Brief Summary Text (34):
wherein in the course of the koji preparation and the proteolysis, removal of phytic acid is advanced in parallel with the separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop to form isoflavone compounds containing aglycones in a large amount, thereby obtaining a product from the pulse crop as a starting material.

Brief Summary Text (35):
Still further, the food of the present invention containing a product prepared from a pulse crop as a starting material contains a product prepared from a pulse crop as a starting material by steps of:

Brief Summary Text (36):
inoculating a koji starter on a pulse crop to prepare koji; and

Brief Summary Text (37):
adding water to the resultant from the koji preparation treatment to advance hydrolysis of a protein contained in the resultant;

Brief Summary Text (38):
wherein in the course of the koji preparation and the proteolysis, separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop is advanced to form isoflavone compounds containing aglycones in a large amount, thereby obtaining a product from the pulse crop as a starting material.

Brief Summary Text (39):
Further, the food of the present invention containing a product prepared from a pulse crop as a starting material contains a product prepared from a pulse crop as a starting material by steps of:

Brief Summary Text (40):
inoculating a koji starter on a pulse crop to prepare koji; and

Brief Summary Text (41):
adding water to the resultant from the koji preparation treatment to advance hydrolysis of a protein contained in the resultant;

Brief Summary Text (42):
wherein in the course of the koji preparation and the proteolysis, removal of phytic acid is advanced in parallel with the separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop to form isoflavone compounds containing aglycones in a large amount, thereby obtaining a product from the pulse crop as a starting material.

Brief Summary Text (44):
According to the process of the present invention for preparing a product from a pulse crop as a starting material it is realized that koji mold is propagated in koji preparation by inoculating koji starter on a pulse crop to hydrolytically separate glycosidic saccharides from isoflavone compounds contained in the pulse crop, and hydrolysis of a protein contained in the resultant from the koji preparation treatment is advanced by adding water thereto in parallel with further hydrolytic separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop to form isoflavone compounds containing aglycones in a large amount.

Brief Summary Text (45):
According to the process of the present invention for preparing a product from a pulse crop as a starting material it is realized that koji mold is propagated in

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koji preparation by inoculating koji starter on a pulse crop to hydrolytically separate glycosidic saccharides from isoflavone compounds contained in the pulse crop and concurrently therewith to remove phytic acid in the pulse crop, and hydrolysis of a protein contained in the resultant from the koji preparation treatment is advanced by adding water thereto in parallel with further hydrolytic separation of glycosidic saccharides from isoflavone compounds contained in the pulse crop to form isoflavone compounds containing aglycones in a large amount and in parallel with further removal of phytic acid.

Brief Summary Text (46):
The koji mold as mentioned above produces various enzymes, and of these enzymes, .beta.-glucosidase, phytase, phosphatase and protease are utilized to hydrolytically separate glycosidic saccharides from isoflavone compounds and to remove phytic acid contained in a pulse crop.

Brief Summary Text (47):
The food of the present invention containing a product made from a pulse crop is a food containing the above-mentioned product made from a pulse crop. The food contains the product having excellent carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect, immunosuppressive effect and the like, and accordingly, it is capable of maintaining parson's health constantly good when ingested as a health food.

Brief Summary Text (48):
The present invention is constructed and functions as described above, and hence the product prepared in accordance therewith is derived from a pulse crop and is of excellence in carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect, immunosuppressive effect and the like. Further, the product is easy of digestion and yet easy of absorption because it is prepared through proteolysis. Accordingly, the product is nutritionally excellent in terms of protein utilization efficiency. In addition, the product can be used for a food, a livestock feed, an aquacultural feed and the like which may be ingested in a satisfactory amount, because no common salt has been added thereto.

Drawing Description Text (2):
FIG. 1 is a flow chart showing one mode of the process for preparing a product from a pulse crop according to the present invention, which comprises forming aglycones having high pharmacological activities from isoflavone compounds contained in a defatted soybean, and one mode of the process which further comprises concurrently removing phytic acid contained in the defatted soybean.

Drawing Description Text (3):
FIG. 2 is a diagram showing temperature characteristics of a mixture with progress of koji preparation time.

Detailed Description Text (3):
FIG. 1 is a flow chart showing one mode of the process for preparing a product from a pulse crop according to the present invention, which comprises hydrolytically separating glycosidic saccharides from isoflavone compounds contained in a defatted product of soybean which is one of pulse crops to form isoflavone compounds containing aglycones in a large amount in the resulting product, and one mode of the process which further comprises concurrently removing phytic acid contained in the defatted soybean.

Detailed Description Text (5):
Explanation will be given along the procedure in FIG. 1. First, a defatted soybean is cooked. By effecting the cooking, propagation of koji is facilitated. The cooking of the defatted soybean may be conducted batchwise or continuously according to the purpose of preparation or the like.

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Detailed Description Text (6):
After completion of the cooking, the defatted soybean is once cooled to adjust water content in the defatted soybean to a level allowing koji to propagate (for example, 40% by weight).

Detailed Description Text (7):
Incidentally, when a defatted soybean or the like is used as a starting material, the step of cooking may be omitted.

Detailed Description Text (9):
That is, the defatted soybean already cooked is inoculated with a koji starter of a koji mold at a predetermined weight ratio, and mixing is conducted to uniformness.

Detailed Description Text (10):
Then, the mixture is charged into a device for preparing koji and kept in a heated condition at an initial temperature of about 28.degree. to 30.degree. C. for a predetermined period of time to ferment the defatted soybean having a water content as low as 40% by weight with koji, thereby hydrolytically separating glycosidic saccharides from isoflavone compounds contained in the defatted soybean to form aglycones. The koji preparation is continued until an enzyme necessary for hydrolytically separating the glycosidic saccharides from the isoflavone compounds.

Detailed Description Text (11):
In this stage, the koji is propagated on the defatted soybean to produce .beta.-glucosidase which is an enzyme hydrolytically separating a glycosidic saccharide from an isoflavone compound, and by this enzyme, glycosidic saccharides are hydrolytically separated from the isoflavone compounds contained in the defatted soybean to form aglycones of the isoflavones.

Detailed Description Text (12):
As the koji starter for the koji preparation, there may be used those which are used preparation of Japanese traditional fermented foods and tempeh and which are safely used for foods, for example, those classified as Aspergillus genus such as Aspergillus usamii, Aspergillus kawachi, Aspergillus awamori, Aspergillus saitoi, Aspergillus oryzae and Aspergillus niger; and those classified as Rhizopus genus.

Detailed Description Text (13):
The fermentation time depends upon the type of koji mold used. However, it is at least 24 hours and is appropriately selected to be sufficient one for hydrolytically separating glycosidic saccharides from the isoflavone compounds contained in the defatted soybean to satisfactory extent.

Detailed Description Text (14):
The temperature of the mixture in the device for preparing koji changes with time, for example, as shown in FIG. 2, as koji preparation proceeds. That is, the temperature gradually rises until the state of the first agitation (mori) is reached 22 hours after the initiation of the koji preparation, and the temperature slightly falls past the first agitation. Then, the temperature rises again until the stage of the second agitation (Naka) is reached 27 hours after the initiation of the koji preparation. Upon stirring the mixture at the "intermediary", the temperature slightly falls. Then, the temperature rises again until the stage of the third agitation (Shimai) is reached 32 hours after the initiation of the koji preparation. Upon stirring the mixture at the the third agitation (Shimai), the temperature slightly falls. Then, the temperature rises again up to 40 hours after the initiation of the koji preparation. Thereafter, the temperature gradually falls until the koji preparation reaches completion 48 hours after the initiation of the koji preparation.

Detailed Description Text (15):

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Then, water is added to the product resulting from the koji preparation, and the mixture is kept in a heated condition at 30.degree. to 65.degree. C. for a predetermined period of time to hydrolyze protein while sufficiently separating glycosidic saccharides from the isoflavone compounds contained in the defatted soybean by the action of .beta.-glucosidase contained in the product to form aglycones of isoflavones.

Detailed Description Text (16):
With respect to the hydrolysis of the protein, hydrolysis time and hydrolysis temperature are appropriately selected depending upon the type of koji used so that glycosidic saccharides are separated from the isoflavone compounds contained in the defatted soybean to satisfactory extent.

Detailed Description Text (17):
In this manner, organic acids are formed in the initial stage of the fermentation to inhibit contaminants in the defatted soybean from propagating, thereby eliminating undesired possibility of secondary contamination. Consequently, a product made from a defatted soybean as a starting material can be mass-produced. Further, even if the water content is not low, it is possible to carry out such treatment for separating glycosidic saccharides from the isoflavone compounds sufficiently.

Detailed Description Text (18):
Table 2 shows contents of isoflavone compounds in 100 g of a defatted soybean which is prepared by subjecting an untreated defatted soybean to koji preparation initiated at an initial temperature of 30.degree. C. and completed over a period of 48 hours, adding water to the resulting product in the same weight as that of the resulting product, and subjecting the mixture to hydrolysis of proteins at 30.degree. C. for 24 hours.

Detailed Description Text (19):
According to Table 2, daidzein and genistein which are aglycones of isoflavone compounds are contained in greatly increased amounts of 74 mg and 59 mg which are about 23 times and 14 times as large as the amounts thereof in the conventional example shown in Table 1, respectively. From this, it is understood that daidzein and genistein can be formed in further increased amounts by effecting the hydrolysis of proteins for 24 hours or more after the completion of the koji preparation.

Detailed Description Text (20):
In another Example, the treatment according to the process of the present invention was applied to an untreated defatted soybean and a soy protein isolate, and Table 3 comparatively shows, for the same purpose as that of Table 2, measurements thereon prior and posterior to the treatment.

Detailed Description Text (21):
Explanation is first made with respect to one of them, the defatted soybean. Proportions of starting materials and koji starter was such that 100 g of a defatted soybean, 0.1 g of a roughly polished rice, and 8.times.10.sup.7 koji spores/g were used. With such proportions, the untreated defatted soybean was subjected to koji preparation initiated at an initial temperature of 30.degree. C. and completed over a period of 48 hours, and water was added to the resulting product in the same weight as that of the resulting product, and the mixture was subjected to hydrolysis of proteins at 50.degree. C. for 48 hours. The results are as shown in Table 3.

Detailed Description Text (22):
As the other of them, i.e., the commercially available soy protein isolate, Fujinic 200 (trade name) manufactured by Fuji-Purina k.K. was used. Proportions of starting materials and koji starter was such that 100 g of the commercially available soy

protein, 0.1 g of a roughly polished rice, and 8.times.10.sup.7 koji spores/g were used. With such proportions, the untreated commercially available soybean protein was subjected to koji preparation initiated at an initial temperature of 30.degree. C. and completed over a period of 48 hours, and water was added to the resulting product in the same weight as that of the resulting product, and the mixture was subjected to hydrolysis of proteins at 50.degree. C. for 48 hours. The results are as shown in Table 3. Table 3

Detailed Description Text (24):
Likewise, in the commercially available soybean protein, daidzein and genistein which are aglycones of isoflavone compounds are contained in greatly increased post-treatment amounts of 100 mg and 94 mg which are about 19 times and 21 times as large as the pre-treatment values, respectively. In addition, the amounts of daidzin and genistin which are isoflavone compounds each having a glycosidic saccharide are extremely reduced to a level as low as 1.0 mg and 3.3 mg, respectively.

Detailed Description Text (27):
The preparation procedure of the invention is conducted in substantially the same manner as in the previously described preparation procedure. However, in the koji preparation step, water addition step and hydrolysis step, phytic acid is removed from the defatted soybean in parallel with the formation of isoflavone compounds containing aglycones in a large amount.

Detailed Description Text (29):
In the koji preparation step, a mixture of a defatted soybean and koji starter is charged into a device for preparing koji and kept in a heated condition at an initial temperature of about 28.degree. to 30.degree. C. for a predetermined period of time to ferment the defatted soybean having water content as low as 40% by weight by means of koji starter until phytic acid in the defatted soybean is sufficiently removed.

Detailed Description Text (30):
In this case, koji mold is propagated on the defatted soybean to produce phytase and phosphatase which are enzymes decomposing phytic acid, and by the enzymes, phytic acid in the defatted soybean is hydrolytically removed.

Detailed Description Text (32):
As the koji starter for the koji preparation, there may be used koji molds which are used preparation of Japanese traditional fermented foods and tempeh and which are safely used for foods, for example, those having high phytase and phosphatase potency and classified as Aspergillus genus such as Aspergillus usamii, Aspergillus kawachi, Aspergillus awamori, Aspergillus saitoi, Aspergillus oryzae and Aspergillus niger; and those having high phytase and phosphatase potency and classified as Rhizopus genus.

Detailed Description Text (33):
The fermentation time depends upon the type of koji mold used. However, it is at least 24 hours and is appropriately selected to be sufficient one for removing phytic acid contained in the defatted soybean to satisfactory extent.

Detailed Description Text (34):
In the subsequent water addition step and hydrolysis step, water is added to the product resulting from the koji preparation, and the mixture is kept in a heated condition at 30.degree. to 55.degree. C. for a predetermined period of time to hydrolyze protein while sufficiently reducing the amount of phytic acid contained in the defatted soybean by the hydrolytic action of phytase, phosphatase and/or protease contained in the product.

Detailed Description Text (35):

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With respect to the hydrolysis of protein, hydrolysis time and hydrolysis temperature are appropriately selected depending upon the type of koji used so that phytic acid contained in the defatted soybean is sufficiently removed.

Detailed Description Text (38):
It is, therefore, preferred to effect removal of phytic acid by liberating at least two phosphoric acid groups from phytic acid which is inositol hexaphosphate to form inositol tetraphosphate, inositol triphosphate, inositol diphosphate, inositol monophosphate or inositol alone or a mixture thereof, thereby obtaining a product which enables minerals to be absorbed efficiently. In this case, it is preferred to control the number of the phosphoric acid groups liberated from phytic acid by adjusting the fermentation time, and hydrolysis time and hydrolysis temperature depending upon the type, state, properties and amount of the pulse crop, the type, state, properties and amount of the koji, and type and properties of the intended product.

Detailed Description Text (39):
Table 4 shows phytic acid content in 100 g of a defatted soybean, with respect to an untreated defatted soybean; defatted soybeans A and B which are prepared using two different shochu kojis (Aspergillus niger and Aspergillus awamori) and each prepared by subjecting a defatted soybean to koji preparation initiated at an initial temperature 30.degree. C. and completed over a period of 48 hours, adding water to the resulting product in the same weight as that of the resulting product, and subjecting the mixture to hydrolysis of protein at 30.degree. C. for 24 hours; and a defatted soybean subjected to conventional washing treatment with an alcohol.

Detailed Description Text (40):
According to Table 4, in contrast to the phytic acid content of 999 mg (about 1%) in the untreated defatted soybean, no substantial phytic acid contents in the defatted soybeans A and B are detected, which are each prepared according to the present invention by subjecting an defatted soybean to shochu koji treatment, adding water to the resulting product in the same weight as that of the resulting product, and subjecting the mixture to hydrolysis of proteins at 30.degree. C. for 24 hours. In other words, almost all phytic acid is decomposed in each of the defatted soybeans A and B.

Detailed Description Text (43):
Next, a food containing the product made from a pulse crop according to the present invention will be described.

Detailed Description Text (44):
The food containing the product made from a pulse crop according to the present invention includes a food consisting only of the product made from a pulse crop which is prepared in accordance with the process of the present invention and a food containing the product in part.

Detailed Description Text (45):
The product made from a pulse crop as a starting material which is prepared in accordance with the process of the present invention is a food having an extremely low salinity, because it is prepared without being salified with common salt. Accordingly, the product can be ingested in a sufficient amount when served as a food. And yet, the food contains aglycones of isoflavones in a large amount, which exhibit excellent carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect and immunosuppressive effect, thereby enabling dietarily desired life to be realized which exhibits excellent effect in terms of health maintenance of a human being.

Detailed Description Text (46):
For example, when the food containing the product made from a pulse crop according

to the present invention is formed into a form convenient for eating such as a biscuit, cookie or the like, it is possible to ingest aglycones of isoflavones which have excellent carcinopreventive and carcinostatic activities, osteoprosis therapeutic effect and immunosuppressive effect while such an article is eaten as a food. In particular, by simply eating such a biscuit or the like in an amount covering the intake of aglycones of isoflavones per day which is required to attain carcinopreventive and carcinostatic effect, osteoprosis therapeutic effect and immunosuppressive effect, the biscuit or the like contributes to prevention of outbreak of the disorders.

Detailed Description Text (47):
Of these aglycones of isoflavones, genistein is effective for prevention and carcinostasis at an initial stage of mastocarcinoma, prostatitic cancer and the like. Accordingly, ingestion of the food containing the product made from a pulse crop according to the present invention contributes to prevention of outbreak of these cancers, thereby enabling dietarily desired life in terms of health maintenance to be realized.

Detailed Description Text (48):
Further, with respect to osteoprosis, while aglycones of isoflavones exhibit osteopenia preventive effect, the removal of phytic acid enables a vitamin B complex having growth promoting activities and antiadipohepatic activities and the like to be maintained highly active and hence exhibits facilitative effect on absorption of calcium contained in the pulse crop. Moreover, these effects synergestically provide a food having extremely excellent osteoprosis therapeutic effect. In particular, such a food exhibits significant effect when used in dietotherapy for a person hormone-relatedly susceptible to osteoprosis.

Detailed Description Text (49):
When the defatted soybean prepared in accordance with the above-described procedure is utilized as a feed or the like, as shown in FIG. 1, the defatted soybean prepared as in the above-described embodiments is dried and then pulverized to obtain a product as a pulverized defatted soybean having high pharmacological activities, such as a material for a livestock feed, an aquacultural feed or the like.

Detailed Description Text (50):
According to the present invention, formation of aglycones of isoflavone compounds contained in a pulse crop, which have high pharmacological activities, at an extremely high formation ratio; removal of phytic acid in the pulse crop; and hydrolysis of proteins are effected by propagation of living koji. Therefore, the formation of aglycones and the removal of phytic acid can be attained even if the pulse crop is in solid state or fluid state, thereby enabling simplified preparation procedure and reduced preparation cost to be realized.

Detailed Description Text (51):
Further, the preparation process of the present invention can be carried out using a conventional device for preparing koji without any alteration, and hence a basic device for production is not required to be specially manufactured, thereby providing wide utility.

Detailed Description Paragraph Table (2):
TABLE 3 commercially avail- defatted soybean
able soybean protein pre post pre post
daizin 100 not detected 90 1.0 daizein 3.2 70 5.3 100 genistin 120 1.3 120 3.3
genistein 4.2 64 4.4 94 (unit: mg/100g)

Detailed Description Paragraph Table (3):
TABLE 4 defatted soybean phytic acid content
(mg/100g) untreated defatted soybean 999

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(mg/100g) shochu koji-treated d. s. A not detected shochu koji-treated d. s. B not detected alcohol-washed d. s. 1,150 (mg/100g)
(detection limit: 5mg/100g)

CLAIMS:

1. A process for preparing a product from a pulse crop as a starting material which comprises the ordered steps of:

preparing a koji preparation by the steps comprising:

cooking said pulse crop,

cooling said cooked pulse crop,

adding water into said pulse crop,

mixing a koji starter into said pulse crop,

incubating said pulse crop while stirring, and

hydrolyzing said koji preparation by adding water, whereby phytic acid contained in said pulse crop is removed and glycosidic saccharides contained in said pulse crop are hydrolyzed, thereby forming isoflavone compounds containing aglycones.

2. A process according to claim 1, wherein said pulse crop is kept between 30.degree.-40.degree. C. during said step of preparing a koji preparation.

3. A process according to claim 1, wherein said glycosidic saccharides contained in said pulse crop are converted into aglycones.

4. A process according to claim 1, wherein said koji starter comprises Aspergillus.

5. A process according to claim 1, wherein said step of hydrolyzing said koji preparation comprises adding a quantity of water approximately equal in weight to the weight of said koji preparation.

6. A process for preparing a product in a bulk quantity from a pulse crop as a starting material which comprises the ordered steps of:

preparing a koji preparation by the steps comprising:

cooking said pulse crop,

cooling said cooked pulse crop,

adding water into said pulse crop,

mixing a koji starter into said pulse crop,

incubating said pulse crop while stirring, and

hydrolyzing said koji preparation by adding a quantity of water approximately equal in weight to the weight of said koji preparation and keeping said koji preparation at 30.degree.-65.degree. C. for approximately 48 hours, whereby phytic acid contained in said pulse crop is removed and glycosidic saccharides contained in isoflavones are converted into aglycones.

7. A food product prepared from a pulse crop as a starting material, said product

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being prepared by the ordered steps of:

preparing a koji preparation by the steps comprising:

cooking said pulse crop,

cooling said cooked pulse crop,

adding water into said pulse crop,

mixing a koji starter into said pulse crop,

incubating said pulse crop while stirring, and

hydrolyzing said koji preparation by adding water, whereby phytic acid contained in said pulse crop is removed and glycosidic saccharides contained in said pulse crop are hydrolyzed, thereby forming isoflavone compounds containing aglycones.

8. A food product prepared from a pulse crop as a starting material, said product being prepared by the ordered steps of:

preparing a koji preparation by the steps comprising:

cooking said pulse crop,

cooling said cooked pulse crop,

adding water into said pulse crop,

mixing a koji starter into said pulse crop, and

incubating said pulse crop while stirring;

hydrolyzing said koji pulse crop by adding a quantity of water approximately equal in weight to the weight of said koji preparation and keeping said koji preparation at 30.degree.-65.degree. C. for approximately 48 hours, whereby phytic acid contained in said pulse crop is removed and glycosidic saccharides contained in isoflavones are converted into aglycones.

9. A food product prepared from a pulse crop as a starting material according to claim 8, further comprising a step of drying said koji pulse crop after said hydrolyzing step.

10. A food product prepared from a pulse crop as a starting material according to claim 9, further comprising a step of pulverizing said koji pulse crop after said drying step.

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